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BACTERICIDAL EFFECTS OF ALGAE ON ENTERIC ORGANISMS

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27 **Abstract**

Various enteric organisms were exposed to axenic cultures of green and blue-green algae to determine what effect the algae might have. Tests were extended to 90 days to determine whether aftergrowth was possible. Mixed cultures of both green and blue-green algae were exposed to both single species and mixed cultures of enteric bacteria, at varying stages of the algae growth periods. Results indicated that mixed cultures do have a pronounced bactericidal effect on enteric organisms, while for single algal species, the effect is less pronounced. Also, the bactericidal effects are more specific for pathogens, with virtually no aftergrowth of pathogens detected. (Lowry-Texas)

(WRSIC abstract)

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BACTERICIDAL EFFECTS OF ALGAE
ON ENTERIC ORGANISMS

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for the

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ABSTRACT

A series of experiments involving the effects of blue-green and green algae on the dieoff rates of selected bacteria have been conducted. The algae were axenic cultures of *Anabaena cylindrica*, *Anacystis nidulans*, *Gloeocapsa alpicola*, *Oscillatoria chalybia*, *O. formosa*, *Phormidium faveolarum*, *Ankistrodesmus braunii*, *Chlorella pyrenoidosa*, *C. vulgaris*, and *Scenedesmus obliquus*. Cultures of enteric bacteria species (*Alcaligenes faecalis*, *Enterobacter aerogenes*, *Escherichia coli*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, and *Serratia marcescens*) were added to the axenic algal cultures during different periods of the algal life cycles.

Cultures of the normal blue-green contaminants were exposed to the enterics to determine antagonistic effects toward the enterics. Filtrate from actively growing algae was exposed to cultures of enterics to determine whether any antibiotic compounds were imparted to the medium during lag phase growth of algae. To determine aftergrowth of the enteric species, the duration of the tests was extended to about 90 days. Mixed cultures of green and blue-green algae were exposed to both single species of enteric bacteria and mixed cultures. The results indicated that mixed algal cultures cause a greater dieoff among the enteric bacteria than do individual species of algae. The dieoff characteristics of pathogenic species, namely, *Salmonella typhosa*, *S. paratyphi*, *Shigella dysenteriae*, *S. paradysenteriae*, and *Vibrio comma* were also determined.

The pathogenic species did not survive as long as the enteric test species under similar test conditions. Virtually no aftergrowth was detected on the part of the pathogens.

CONCLUSIONS

The following conclusions are derived from the results of this investigation.

1. Dieoff coefficients for individual species of enteric bacteria in the presence of axenic cultures of algae were comparatively low, the majority of the coefficients being near or less than -0.1 per day. *Chlorella pyrenoidosa* and *Chlorella vulgaris* caused the highest dieoff coefficients among enteric bacteria. *Chlorella spp*. were substantially more effective than *Ankistrodesmus braunii* or *Scenedesmus obliquus* in effecting accelerated dieoffs.

2. Mixed cultures of either the blue-green or green algae caused significantly higher dieoff coefficients among the enteric test bacteria as well as the pathogenic bacteria tested. The majority of the coefficients were between -0.1 and -0.2 per day.

3. Effects exhibited by enteric bacteria on the growth of individual algal species depended on the algal species in question. Constant patterns of increased or decreased algal growth coefficients were uncommon. In the majority of algae species, a slight inhibition of the overall growth potential of the algae was observed.

4. Dieoff of enteric bacteria was more rapid under aerobic conditions than anaerobic conditions.

5. Aftergrowth of *Escherichia coli*, *Pseudomonas aeruginosa*, and *Serratia marcescens* occurred in axenic blue-green algal cultures as well as in waste stabilization pond effluent. *Alcaligenes faecalis*, *Enterobacter aerogenes*, *Proteus vulgaris*, *Vibrio comma*, *Salmonella typhosa*, *Salmonella paratyphi*, *Shigella paradysenteriae* exhibited no aftergrowth potential under similar conditions. *Serratia* and *Pseudomonas* exhibited a greater aftergrowth potential than did *E. coli*.

6. As the algal species reached their stationary and/or log death-growth phase in the laboratory, quantities of organic carbon were released to the medium; up to 200 mg/l was not uncommon. Prolonged survival periods and/or aftergrowth by some of the enteric bacteria were attributed to this nutrient source.

7. Consistent dieoff effects on enteric bacteria in laboratory and field waste stabilization ponds were achieved only after appropriate periods of acclimitization of the pond microcosms. Those periods were observed to be as long as 30 days, or more in some instances. Dieoff coefficients for early stages in pond treatment units were higher than those obtained for secondary stages such as maturation ponds. Higher coliform concentrations and increased competition for nutrient sources in early treatment sequences were attributed to that rapid dieoff.

8. Compared to axenic algal culture experiments and laboratory scale ponds, the most rapid reduction in enteric bacteria occurred in the waste stabilization ponds located in the field.

9. In laboratory ponds, E. coli exhibited a greater resistance to dieoff than did Pseudomonas aeruginosa or Serratia marcescens; but in the field ponds, E. coli exhibited the highest rate of dieoff of any enteric bacterial species tested.

10. Occasional increases in concentrations of Pseudomonas and Serratia were noted in laboratory and field ponds. Short-circuiting was not considered to be the causative factor, but an association of these two genera and other enteric bacteria with clumps of algae might have been responsible for this increase. Pseudomonas spp. exhibited increases in numbers when the total algal concentrations were lowest in both the laboratory and field ponds.

11. Total coliform bacteria counts decreased significantly during periods when the pond phytoplankton population was highest, and vice versa.

12. The vast majority of bacteria in all pond effluents were of the group of bacteria referred to as the chromagens; included in the group are Flaveobacterium and Brevibacterium. Cultures of these two separate

genera were shown to exert marked antagonistic effects on enteric bacteria when together in culture. Flaveobacterium was more antagonistic to enteric bacterial species than Brevibacterium.

13. On several occasions extended periods of incubation were necessary to produce any recordable growth of Pseudomonas spp. from waste stabilization pond samples using either nutrient, trypticase soy, or Endo agar plates. Special consideration should be given this factor when total or enteric counts are made from wastewater environments.

TABLE OF CONTENTS

PAGE

ACKNOWLEDGMENTS iii

ABSTRACT iv

CONCLUSIONS v

TABLE OF CONTENTS viii

LIST OF TABLES ix

LIST OF FIGURES x

CHAPTER

1 INTRODUCTION 1
  - Bacterial Characteristics 1
  - Review of Literature 3

2 MATERIALS AND METHODS 9
  - Algal and Bacterial Cultures 9
  - Laboratory Investigation Series Identification 13
  - Laboratory Data Analyses Methods 15
  - Laboratory and Field Waste Stabilization Pond Studies 16

3 LABORATORY CULTURE DIEOFF EXPERIMENTS 17
  - Enteric Bacteria Dieoff Studies 18
  - Pathogenic Bacteria Dieoff Studies 25
  - Aftergrowth Potential Measurements 25
  - Organic Carbon Production by Algae 27

4 LABORATORY AND FIELD WASTE STABILIZATION POND STUDIES 30
  - Laboratory Waste Stabilization Pond Studies 30
  - Field Waste Stabilization Pond Studies 44

BIBLIOGRAPHY 54

APPENDIX A: STATISTICS OF ALL LABORATORY AXENIC CULTURE STUDIES 59

APPENDIX B: BACTERIOLOGICAL DATA FROM LABORATORY AND FIELD WASTE STABILIZATION POND STUDIES 97

APPENDIX C: PROGRAM BETA FORMAT 126

LIST OF TABLES

TABLE		PAGE
2-1	Composition of Algal Culture Medium	10
2-2	Element Concentrations in Algal Culture Medium	11
2-3	Algae and Bacteria Test Species	12
3-1	Dieoff Coefficients for Series Utilizing Axenic Algal Cultures and Enteric Bacteria	19
3-2	Dieoff Coefficients for Series Utilizing Pathogenic Bacterial Species	26
3-3	Aftergrowth Characteristics of Enteric Bacterial Species with Single Species of Algae (Series I)	28
3-4	Total Carbon and Total Organic Carbon Content of Biomass After Ninety Days (Series BG-I, G-I, BG-VIII, and G-VIII, in mg/l)	29
4-1	Bacteria Inoculated Into Selected Stations In Laboratory Waste Stabilization Ponds	33
4-2	Dieoff Coefficients of Inoculated Bacteria in Laboratory Scale Waste Stabilization Ponds	34
4-3	Phytoplankton Found in Laboratory Waste Stabilization Ponds	36
4-4	Total Phytoplankton Concentrations Found In Laboratory Waste Stabilization Ponds	37
4-5	Bacteria Inoculated Into Selected Stations of Waste Stabilization Ponds	46
4-6	Phytoplankton Found in Waste Stabilization Ponds	51
4-7	Phytoplankton Concentrations Foundin Waste Stabilization Ponds	52
A-1 through A-72		
	Appendix A: Statistics of All Laboratory Axenic Culture Studies	59
B-1 through B-28		
	Appendix B: Bacteriological Data from Laboratory and Field Waste Stabilization Pond Studies	97

LIST OF FIGURES

FIGURE		PAGE
4-1	Schematic of Laboratory Ponds	31
4-2	Total Bacteria, Laboratory Ponds	38
4-3	Total Coliform, Laboratory Ponds	39
4-4	_E. coli_, Laboratory Ponds	40
4-5	Phytoplankton Densities in Laboratory Ponds	41
4-6	Schematic of Waste Stabilization Ponds	45
4-7	Total Bacteria in Waste Stabilization Ponds	48
4-8	Total Coliform in Waste Stabilization Ponds	49
4-9	Phytoplankton Densities in Waste Stabilization Ponds	50

importance. Alcaligenes faecalis has been isolated from infections of bacteremias, gall bladder infections, eye infections, and has frequently been incriminated in cases of enteritis. Species of Pseudomonas, by the same token, are frequently encountered in eye and ear infections as well as urinary tract infections (26). This genus is universally treated with a great deal of respect, especially in facilities in which burn patients are housed. A septicemia caused by Pseudomonas may occur as frequently as staphylococcal septicemia in severely burned patients and in persons who have leukemia. The outcome is usually fatal.

The genus Vibrio, which has several nonpathogenic water-borne species, is also found in the Family Pseudomonadaceae. Most dangerous of the species are Vibrio comma and Vibrio El tor which are the causative agents of the well-known Asiatic cholera.

Review of Literature

The mechanism by which populations of undesirable bacteria are reduced in numbers has been the subject of many investigations. In waste treatment facilities the bacterial dieoff is affected by several factors. In lakes, reservoirs, impoundments, and streams the bacterial dieoff may be assumed to be similar insofar as these factors are concerned. The principal difference between the aquatic environments is one of bacterial concentration. Some of the factors which undoubtedly play an important part in the bacterial dieoff mechanism are sunlight, pH changes, changes in oxygen tension, predation by other organisms such as rotifers, changes in organic content of the water, temperature, and antagonistic effects of other bacterial species and other faunistic species such as fungi and algae. Gravel, et al. (2) found temperature, pH, and dissolved oxygen concentration to be important, in that order, in dieoff rates of reservoir coliforms. Gameson and Saxon (3) attributed the dieoff primarily to sunlight effects.

Bacteria must have certain quantities of organic carbon present for their survival or multiplication. Ward and Moyer (14) reported that organics excreted by algae during growth could serve as bacterial nutrient sources. This source of carbon may reach appreciable concentration levels. Hellebust (13) reported that some phytoplankton are capable of excreting up to 25 percent of their photoassimilated carbon during their log growth phase. Therefore, when large populations of algae are present, adequate supplies of carbon should be present for the survival of some of the coliform bacteria. Data presented by McGrew and Mallette (10) stated that some bacteria of intestinal origin, including *Escherichia coli*, could survive and even multiply at concentration levels of glucose less than 5 micrograms per milliliter.

The literature contains various reports of interactions between coliforms and other faunistic species such as algae. McLachlan and Yentsch (17) and Nakamura (18), respectively, found that certain bacteria enhanced the growth of *Dunaliella* and *Chlorella*. Ward and Moyer (14) and later Ward, Moyer, and Vela (25) demonstrated that there was significant reduction in growth of *Chlorella pyrenoidosa* when in the presence of *Pseudomonas aeruginosa*. Opposing opinions can be found regarding the antagonism of microorganisms to one another. Guthrie *et al.* (11) and Geldreich and Clarke (4) have identified inhibition characteristics between *Pseudomonas aeruginosa* and *Escherichia coli* under different environmental conditions.

The interactions which occur between bacteria and algae may affect the physiology and productivity of an aquatic community. Stimulation of bacteria by algae or algal exudates has been reported by the following investigators. Recent work by Vela and Guerra (52) and Ward, *et al.* (25) furnished evidence that, in some cases, the proliferation of bacterial species may be a function of algal growth. In tests involving *Shigella*, *Proteus*, *Staphylococcus*, *Streptococcus*, and *Corynebacterium* they found rapid dieoff patterns of these bacteria when exposed to *Chlorella*. Yet, it was also reported that *Salmonella typhi* and *Salmonella paratyphi* grew well in the presence of *Chlorella*. In extensive works on toxic blooms of blue-green algae, Gorham (36) found that *Microcystis* produced a toxin but stated that it did not inhibit bacteria such as *Bacillus subtilis*, *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas hydrophila*. It was hypothesized that the age of the culture may have been an important factor in the results and should be considered when analyzing future data. Fogg (15, 16) also reported stimulatory effects to bacteria when associated with algae. He attributed those effects to extracellular by-products of the algae. Lefevre (37) substantiated earlier hypotheses that the extracellular products did exist.

The numbers of reports which conclude that algae may be inhibitory to bacterial growth are in the majority. Geldreich and Clarke (4) reported that fecal coliform bacteria were influenced adversely by Schizothrix calcicola within 24 hours. Telutchenko and Fodorov (5) concluded that their algae affected the test bacteria by using the carbon dioxide and, thus, shifting the pH, by releasing antibacterial substances, by inhibiting the bacteriophage which lyse bacteria, and by increasing the organic content which stimulated the growth of the bacteria. They further concluded that Chlorella vulgaris was more efficient than Scenedesmus obliquus in killing E. coli and Salmonella typhimurium. Chlorella vulgaris was the test algal species used by Pratt and Fong (19). Their conclusion was that that species of Chlorella was capable of inhibiting the growth of associated bacteria.

Birge and Judey in 1929 (28) indicated that algae may have a role in reducing the numbers of bacteria in water. To date only "Chlorellin" has been named specifically with regard to its antibacterial characteristic by Caldwell (30), Pratt (46), and Spoehr, et al. (50). Flint and Moreland (35) were able to demonstrate that metabolic exudates of certain blue-green algae were toxic to bacteria but carried the report no further. Neel and Hopkins (43) observed the reduction in numbers of types of coliforms during seasons of the year in which prolific algal growth occurred in the ponds. Work by Vladimirava (53) reported that cultures of Chlorella pyrenoidosa were definitely capable of suppressing bacterial growth. Prescott (47) cited two genera, Microcystis and Chlorella, as being capable of producing and secreting substances active against two bacterial genera, Staphylococcus and Clostridium. Oswald and Gotaas (45), in an extensive work dealing with pilot-plant waste stabilization ponds, proposed that no specific anticoliform activity could be credited to an algal culture tested in the laboratory. However, they did not discount the possibility of antibacterial properties of algae.

The problem of bacterial contamination has been overlooked by some investigators and this is of importance when testing single species of algae. It is doubtful that there are many bacteria-free cultures of filamentous blue-green or green algae. Unicellular or diplo forms of blue-greens and greens are relatively easy to grow in a bacteria-free state. In naturally contaminated cultures, Ward and Moyer (14) reported that the bacterial mass was less than one percent of the algal mass. Yet, the numbers of bacteria were shown to exceed a million per milliliter; figures approaching a billion per milliliter were not uncommon. As to the contaminants themselves, Krauss and Thomas (20) reported Flavobacterium to be the most common and persistent genus in cultures of Scenedesmus obliquus. Levinson and Tew (21) also reported Flaveobacterium as a contaminant of their research cultures of algae. Their test species was Chlorella vulgaris.

Numerous reports are available which quote reductions in the coliform numbers through waste stabilization ponds. The reduction percentages are usually impressive; however, as Geldreich (1) pointed out, even with reductions of from 90-99 percent the remaining 1-10 percent of the coliforms may easily constitute numbers of from 4×10^6 to 10×10^6 per 100 milliliters. These values are not acceptable for more effluent standards. Most of the species incorporated in the coliform group obviously have a similar metabolic pattern of growth and development. Geldreich (7) and Gallagher and Spino (8) have observed similar survival characteristics (or death rates as the case may be) among the more abundant coliform species. In making observations on streams, Churchill (9) reported that the slopes of die-away curves for the total and fecal coliforms were essentially the same. While figures are usually the best measure of coliform dieoff when describing their functions, it must be remembered that a rate number does not relate the environmental conditions. In reports dealing with enteric bacteria reduction in ponds and series of ponds in South Africa and Zambia, Marais and Shaw (40), and later Marais (38, 39) used a value of $K = 2.0$ for E. coli and $K = 0.8$ for Salmonella typhi. These differences between only two species indicate the need for further data. Projections made by use of the modifications of Chick's Law as reported by Marais (38, 39)

CHAPTER 1

INTRODUCTION

An important reason for the treatment of domestic wastewaters is the reduction or elimination of the enteric bacteria from these wastewaters; in this connection, waste stabilization ponds have been used successfully. In established ponds the most obvious population consists of various species of algae as evidenced by their pronounced color. These algae, under proper pond design, can produce the greater percentage of required dissolved oxygen and can interact with the entire biological community. As yet, however, the specific role that algae play in the overall reduction of enteric bacteria in waste stabilization pond systems has not been firmly established.

The purpose of this investigation was to determine the degree of toxicity exerted by typical species of blue-green and green algae on representative bacteria found in wastewaters. The scope of this investigation included: (a) long-term studies involving selected species of algae, coliform bacteria, and pathogenic bacteria; (b) bactericidal and bacteristatic effects; (c) algal culture filtrate effects on bacteria test species; (d) aftergrowth capabilities of test bacteria by extension of study periods; and (e) enteric bacteria dieoff investigations in both laboratory waste stabilization ponds and field ponds having different design characteristics.

Bacterial Characteristics

A brief description of some of the important characteristics of the bacteria studied in this investigation is appropriate in order that their complexity and the significance of their reduction in wastewaters can be fully appreciated. Coliform bacteria are, by definition and description, inclusive of all aerobic and facultative anaerobic, gram-negative, nonsporeforming rod shaped bacteria which are capable of fermenting lactose with gas formation within forty-eight hours at a temperature of 35 degrees

Centrigrade (27). They have been variously named the B. coli group and the coli-aerogenes group in past years with no change in the specifications. By this definition, therefore, only part of the bacterial flora inhabiting the gastrointestinal tract of animals are coliforms. The total number of genera which are capable of living in those conditions is largely unknown. Invariably, species and varieties of Escherichia, Streptococcus, Clostridium, Aerobacter (renamed Enterobacter), Paracolobactrum, Salmonella, Shigella, Proteus, Pseudomonas, Alcaligenes, Serratia, and Bacteriodes are among those found. In this sense any species which has the capability to survive and multiply in any intestinal tract could be called an enteric bacterium. Taxonomically, the enteric bacteria follow this classification (26):

- Order Eubacteriales
  - Family Enterobacteriaceae
    - Genus Escherichia
    - Genus Aerobacter
    - Genus Klebsiella
    - Genus Paracolobactrum
    - Genus Alginobacter
    - Genus Erwinia
    - Genus Serratia
    - Genus Proteus
    - Genus Salmonella
    - Genus Shigella

Other genera which are found routinely in domestic wastewaters have the following classification:

- Order Eubacteriales
  - Family Achromobacteraceae
    - Genus Alcaligenes
- Order Pseudomonadales
  - Family Pseudomonadaceae
    - Genus Pseudomonas

To assume that all of the species of the genera listed in the above classifications are nonpathogenic would be erroneous. Several species in the Family Enterobacteriaceae, for example, have been known to be pathogenic to man, producing various intestinal diseases and septicemic infections. For this reason alone their elimination from wastewaters is of utmost may be used effectively to provide an insight into what general bacterial pollution control may be required. Hanes, et al. (6) reported log death rates of 0.134/day, 0.291/day, and 0.355/day respectively at temperatures of 10°, 20° and 30°.

It is apparent that not enough information is available on pathogenic bacteria such as Salmonella, Shigella, and Vibrio. With large numbers of coliforms present, the probability of finding Salmonella, for example, increases. Of course, with several hundred serotypes of Salmonella in existence, it would be difficult to establish the exact nature of the type found in the sample. Geldreich (12) reported that when the fecal coliform count exceeded 1,000, the Salmonella also increased. Periodic reports of isolation of various species of Salmonella are routine (22). In March 1969, 1,165 isolations of Salmonella were reported for humans, an average of 291 per week. This was an increase of 13.2 percent over the average for February 1969, and an increase of 7.0 percent (weekly) for March 1968. At the same time, 738 non-human isolations occurred during March 1969. These figures indicate that, even through few outbreaks of disease caused by Salmonella are occurring in this country, the causative agents are ever-present. Ward and Moyer (14) reported that Salmonella typhi and Salmonella paratyphi grew well in algal cultures for periods of time extending through seven days. Sidio (48) reported up to 99 percent removal of coliforms along with complete removal of the pathogenic genus, Salmonella. There is additional evidence to indicate that Salmonella typhi survival is dependent on the available supply of nutrients. Increased loadings with shorter retention times were seen to support the survival of the typhoid bacillus. This was reported by McGarry and Bouthillier (41). They also reported that ponds with longer detention times and reduced nutrient concentrations provided a more antagonistic environment. Goetzee and Fourie (31) showed in field studies that waste stabilization ponds operating in series were capable of reducing Salmonella spp. by at least 99.5 percent. The total reduction of E. coli was similar to that found for other bacteria. These investigators reported that Salmonella spp. was more resistant, as compared to E. coli, in highly polluted waters.

Generally, the operating data on waste stabilization ponds are well documented (54, 55, 56, 57, 58, 59). Smallhorst and Walton (49) and Towne, et al. (51) have observed and reported the reduction of enteric organisms in waste stabilization ponds. They attributed this reduction primarily to detention. Towne, et al. (51) also reported that the reduction in coliform numbers was not appreciably different for the seasons despite variations in algal concentrations. Detention coupled with short-circuiting was considered by van Eck (32, 33, 34) and Bolitho (29) to be the most important parameter which influenced bacterial concentration. Reductions in coliform bacteria of above 90 percent routinely occur in ponds which are functioning in an acceptable manner (42, 48, 54). Gann, et al. (23) found Achromobacter 65 percent of the total population of pond bacteria, Pseudomonas 25 percent, Flaveobacterium 5 percent, and the coliforms less than (or rarely equalling) 10 percent.

One aspect of the dieoff of bacteria in treatment facilities which has received very little attention has been the aftergrowth phenomenon. After treatment and discharge, the surviving bacteria, including those which have been exposed to chlorine, may find suitable growth conditions in the receiving waters and continue to multiply. This aftergrowth has been reported by Orlob (44), Geldreich (12), Eliassen (24) and others. Under certain conditions coliform bacteria were found to increase in numbers to peak values within 30 hours up to 10-40 times the original number.(24). Even with chlorination of 15 minutes duration, a lesser increase in aftergrowth occurred of 1-12 times the original number of bacteria. Clearly a greater understanding of the ability of these organisms to reproduce and the accompanhing necessary conditions is needed.

CHAPTER 2

MATERIALS AND METHODS

Algal and Bacterial Cultures

Standard microbiological laboratory procedures were incorporated during all phases of this investigation. Algal species were maintained as bacteria-free as possible under normal laboratory conditions prior to additions of the test bacteria. The axenic algal cultures were subjected to transfer from a solid algal growth medium to a liquid medium, and vice versa, for a period of three years prior to the initiation of the experimentation. Their acclimation to laboratory growth conditions and growth rate constancy was, therefore, assured. The composition of the liquid medium which was used for the culture of the algae in the laboratory is described in Table 2-1. This medium was designed to allow optimum growth of the algae for prolonged periods of time, a feature which was of great benefit during the extended periods of testing necessary for successful completion of the investigation. Table 2-2 presents a breakdown of the elemental concentrations in the algal growth medium shown in Table 2-1.

Throughout the laboratory phase of the investigation six species of blue-green algae and four species of green algae were used as test organisms. These species are listed in Table 2-3 along with the bacteria which were tested. Code numbers for the algae indicate their culture number as cataloged by the culture collection group at Indiana University from where they were obtained. Code numbers for the bacterial species represent the American Type Culture Collection number or the culture number from stock cultures at North Texas State University or both.

The algae were grown in culture and used in tests at a temperature of 25 ± 1°C. Fluorescent lighting operating on a cycle of 14 hours on and 10 hours off provided an intensity of 290-300 foot-candles.

All of the bacterial species were cultured in the laboratory with trypticase soy broth or agar supplemented with 2 g/l yeast extract. Appropriate serial dilutions were made of the cultures followed by counting

Table 2-1. Composition of Algal Culture Medium

Compound	Final Concentration (mg/l)
$NaHCO_3$	200
$MgSO_4$	75
$NaSiO_3 \cdot 5H_2O$	20
$Ca(NO_3)_2$	50
KH_2PO_4	20
NH_4NO_3	75
KNO_3	40

Trace Element Solution (1 ml of the following mixture)

EDTA	10.0 g/l
$ZnSO_4 \cdot 7H_2O$	1.0 g/l
H_3BO_3	1.0 g/l
$MnCl_2 \cdot 4H_2O$	0.5 g/l
$FeSO_4 \cdot 7H_2O$	0.5 g/l
$CoCl_2 \cdot 6H_2Oq$	0.15 g/l
$CuSO_4 \cdot 5H_2O$	0.15 g/l
$(NH_4)_6Mo_7O_{24} \cdot 4H_2O$	0.10 g/l
LiCl	0.0278 g/l
$Al_2(SO_4)_3 \cdot 18H_2O$	0.0556 g/l
$SnCl_2 \cdot 2H_2O$	0.0278 g/l
KI	0.0278 g/l
KBr	0.0278 g/l

Distilled Water, to make 1 liter final volume

The pH of the above medium is adjusted to $\approx$ 5.8 with HCl before autoclaving, resulting in a final pH of $\approx$ 7.4

Table 2-2. Element Concentrations in Algal Culture Medium

Element	Final Concentration (mg/l)
N	41.15
K	21.245
P	4.560
Na	58.468
C	31.825
Cl	102.755
S	20.146
Mg	15.150
Ca	12.200
Si	2.960
Zn	0.228
B	0.174
Mn	0.139
Fe	0.101
Co	0.037
Cu	0.038
Mo	0.054
Li	0.004
Al	0.004
Sn	0.015
I	0.021
Br	0.019

Table 2-3. Algae and Bacteria Test Species

Organism	Code Number
Anabaena cylindrica	B 629
Anacystis nidulans	625
Gloeocapsa alpicola	B 589
Oscillatoria chalybia	B 386
Oscillatoria formosa	LB 390
Phormidium faveolarum	B 427
Ankistrodesmus braunii	245
Chlorella pyrenoidosa	26
Chlorella vulgaris	29
Scenedesmus obliquus	72
Alcaligenes faecalis	ATCC 8748
Enterobacter aerogenes	ATCC 9621
Escherichia coli	ATCC 8677 NT 201
Proteus vulgaris	ATCC 8427
Pseudomonas aeruginosa	ATCC 7700 NT 99
Serratia marcescens	ATCC 13880
Salmonella paratyphi	NT 113
Salmonella typhosa	NT 118
Shigella paradysenteriae	NT 131
Shigella dysenteriae	NT 127
Vibrio comma	NT 154

and enumeration of the bacterial colonies on poured plates of the trypticase soy agar. This mixture of nutrient sources was found to be superior to nutrient broth or nutrient agar alone for growth and subsequent enumeration of most of the test species. In tests involving laboratory cultures of algae and bacteria, counts were made after 24 hours of incubation at 37°C. The plates were then returned to the incubator and recounted at 48 and 72 hours because, in the majority of cases, the algal contaminants and some of the test bacteria did not show adequate growth at 24 hours for accurate enumeration. This recounting technique is time-consuming but necessary.

Two genera of bacteria were the primary contaminants of the filamentous blue-green test algal species. These were Brevibacterium and Flavobacterium and were identified through selective testing procedures by Dr. R. K. Guthrie. Their presence was not detected in the test cultures of green algae.

Addition of test bacterial populations to algal cultures was uniformly controlled throughout the investigation. A standard straight-wire inoculum of the bacterial species in question was incubated for 24 hours in half strength TS broth. The solution was mechanically agitated and 0.05 ml was transferred to each 100 ml of algal culture. Before removing samples from the algal cultures the volume of each 250 ml erlenmeyer flask was adjusted with sterile distilled water to correct for evaporation losses. After making appropriate serological dilutions the samples were plated onto TS agar and counted.

Laboratory Investigation Series Identification

The first phase determined the effects that axenic cultures of blue-green and green algae had on the dieoff of individual species of test bacteria. Additional tests involved studies of algal growth characteristics when exposed to the bacteria and studies of the effects of the contaminating bacteria on various enteric species. The tests are described below.

Series I. Viable bacteria were added to individual algal cultures at controlled times (early to mid-log growth phase of algae) and the bactericidal or bacteristatic effects noted. This series of tests was coded BG-I (blue-greens) and G-I (greens).

Series II.	Viable bacteria were added to individual axenic cultures of algae during the first twenty-four hour period of the lag growth phase after inoculation of the algae into sterile growth medium. Any inhibition of algal growth was determined by this timing sequence. The series was coded BG-II and G-II.
Series III.	Control tests were conducted to evaluate normal cyclic influences of contaminant bacteria in algal cultures. This series was coded BG-III and G-III.
Series IV.	Algal mass was determined by weighing procedures. Comparison of these data from control cycles with data obtained during Series I and II demonstrated whether inhibition or enhancement of algal cultures was the result of the presence of enteric bacteria. This series was coded BG-IV and G-IV.
Series V.	Dieoff rates of the contaminant bacterial species were determined during series involving the additions of enteric bacteria in algal cultures. This series was coded BG-V or "contaminants."
Series VI.	Dieoff rates of the enteric bacteria alone in algal growth medium were analyzed. This series provided the basic control for the study described in Series V above. This series was coded VI.
Series VII.	Separation of the algal growth medium into a cell-free filtrate during mid-log phase of algal growth control with subsequent inoculation of enteric bacteria demonstrated the influences of algal metabolic exudates on the enteric bacteria. This series was coded BG-VII and G-VII.
Series VIII.	Equal quantities of each of the six test species of blue-green algae were mixed when in their mid-log growth phase. Additions of suspensions of individual species of enteric bacteria in each resulting

heterogeneous algal culture demonstrated comparative dieoff rates with Series I and II above. This study was conducted with the blue-greens and the greens separately and was coded BG-VIII and G-VIII.

Series IX. Duplication of series BG-VIII and G-VIII using mixtures of all the enteric bacterial test species provided bacterial dieoff rates which might be expected from field conditions. This series was coded BG-IX and G-IX.

Series X. Testing in the majority of the series was continued for periods up to 90 days to establish patterns of aftergrowth of each bacterial species. This series was coded BG-X and G-X.

Laboratory Data Analyses Methods

Data for all of the series, I through X, and for the runs involving the pathogenic bacteria, were analyzed using a method of least squares. The program (BETA) is shown in Appendix C. It should be noted that when aftergrowth occurred, the data were not subjected to statistical analysis. In order to be acceptable, only decreases in numbers which extended over periods of 90 days were programmed. Data for aftergrowth characteristics are presented separately. Basically, the function of program BETA was the calcultion of the constants for an equation similar to the following:

$$Y = C_0 + C_1 X_1 + C_2 X_2 + C_3 X_3 + \dots + C_n X \qquad 2\text{-}1$$

Y is the dependent variable, X is an independent variable, and C is constant. Equation 2-2 was used to calculate the log (base 10) death rates:

$$\log (Y) = C_0 + C_1 X_1 \qquad 2\text{-}2$$

where

Y = the density of bacteria in number per ml, or mg/l weight

C_0 = the Y-axis intercept

X_1 = the time in days corresponding to Y

C_1 = the death rate coefficient, log base 10

Additional computer output provided: (a) the variance ratio from the horizontal line; (b) variance about the regression line; and (c) the multiple correlation coefficient. Data were analyzed at the 90 percent confidence limit.

Laboratory and Field Waste Stabilization Pond Studies

Additional data were obtained from model waste stabilization ponds. These laboratory units consisted of two serial connected aquaria similar to those described by Malina and Yousef (60). The total capacity was 46 liters. A diagram of these units is shown in Figure 4-1. All of the model pond experiments were conducted at ambient temperature on location at the Govalle Wastewater Treatment Plant in Austin, Texas. Lighting was provided by banks of fluorescent bulbs held at approximately 25 cm above the water surface. The intensity was 325-350 foot-candles during the 12 hours they were cycled.

Three different design concepts were represented by the model ponds. The volumes and detention times were calculated to correspond to the series of full-scale ponds existing at the Govalle facility. The first set of ponds consisted of an anaerobic pond followed by two 46-liter facultative ponds and a maturation pond. The second set was represented by facultative ponds which contained an anaerobic "trench," followed by a maturation pond. The third set contained facultative ponds followed by a maturation pond.

Daily additions of 500 ml untreated domestic wastewater were added to each model pond; a similar quantity was removed from the opposite end to effect a balanced system, and evaporation losses were corrected by the addition of tap water.

CHAPTER 3

LABORATORY CULTURE DIEOFF EXPERIMENTS

Regardless of the degree of laboratory control, considerable variance occurred in the bacterial population during some of the tests. These fluctuations in numbers were mainly attributed to the growth phase of the algal cultures and the inherent nature of bacteria to adhere to filaments or aggregates of algae. The fluctuations also resulted in low multiple correlation coefficients and high variances. All results of the analyses made during the laboratory axenic culture series are presented in Tables A-1 through A-72 of Appendix A. Those data represent the analyses of laboratory data as taken from program BETA printouts.

Column headings in Tables A-1 through A-72 inclusive, Appendix A are as follows:

N = number of data points used in computing that particular regression line

S_H^2 = the variance of data points about the mean of all data points

S_r^2 = the variation of data points about the regression line

S_H^2/S_r^2 = the variance ratio which if referenced against appropriate standard "F" tables would indicate the statistical validity of the data

b = calculated y - intercept

k = the dieoff (-) or growth (+) rate coefficient of the typical $C_t = C_0\ 10^{kt}$ formulation

R = the multiple correlation coefficient

The numbers of data points indicated for each experiment do not necessarily correspond to the total number obtained during the duration of the run. Where significant aftergrowth of test bacterial populations occurred, the test results were not included in the computer program and those data are discussed separately.

Enteric Bacteria Dieoff Studies

Although single species of bacteria do not exist in nature with axenic cultures of algae, without these basic data it would be impossible to assess the true value of each algal species with respect to its effect on the dieoff rate of the bacteria in question. Dieoff rate coefficients for virtually all of the laboratory series involving axenic algal cultures and enteric bacteria are presented in Table 3-1. Considering the series involving the blue-green algal test species, it is easily discernible that no two species exerted the same dieoff effect on any two bacterial species. Additionally, the dieoff rate coefficients vary considerably. One primarily important conclusion which is derived after examination of the data in Table 3-1 is that the comparatively rapid dieoff rates of enteric bacteria which occur in nature are apparently not due to the effects of individual algal species.

Differences in the dieoff rate coefficients (hereinafter called "coefficients") between series BG-I and BG-II were minimal. There was little difference in the effects which algae had on the dieoff rates of the test bacteria when the algae were exposed to the bacteria during algal log growth phase (Series BG-II) or algal log growth phase (Series BG-I). Several of the coefficients seen in Table 3-1 appear with relatively high ranges, as for example, filtrate of Anabaena cylindrica and Alcaligenes faecalis, -.0230 ± .0570 day^{-1} As in this example, some actually exceed the numerical values of the coefficients themselves. It is believed that significant fluctuations occurred in the bacterial populations throughout the experiment duration because of aggregation and adhesive phenomena which are constantly occurring. For these reasons the seemingly high coefficient ranges are not to be regarded as errors.

On the other hand, comparatively low coefficients were due to the abilities of the test bacteria to derive nutritional benefits from the cellular materials of the blue-green algae. One such material was the gelatinous matrix which is a characteristic of all the blue-green algae. Additional evidence of these occurrences can be seen by the coefficients derived from the tests using blue-green algal filtrate. On a comparative basis it appeared that prolonged survival of enteric bacteria occurred when blue-green algae were present as compared to green algae.

Table 3-1. Dieoff Coefficients for Series Utilizing Axenic Algal Cultures and Enteric Bacteria.

Series	Alcaligenes faecalis*	Enterobacter aerogenes*	Escherichia coli*	Proteus vulgaris*	Pseudomonas aeruginosa*	Serratia marcescens*
Anabaena cylindrica						
Mid-log inoc. BG-I	-.0774 ± .0192	-.0854 ± .0107	-.0397 ± .0125	-.1000 ± .0213	-.0512 ± .0241	-.0366 ± .0404
Contaminant redn. BG-I	-.0157 ± .0526	-.0175 ± .0147	-.0173 ± .1320	-.0046 ± .0118	-.0154 ± .0066	-.0027 ± .0072
Day-0 inoc. BG-II	-.0582 ± .0419	-.0816 ± .0557	-.1132 ± .0534	-.0687 ± .0307	-.0702 ± .0104	-.0583 ± .0249
Contaminant redn. BG-II	-.0187 ± .0054	-.0159 ± .0128	-.0175 ± .0258	-.0094 ± .0215	-.0198 ± .0163	-.0234 ± .0157
Filtrate BG-VII	-.0230 ± .0570	-.0316 ± .0832	-.0164 ± .0575	-.0253 ± .1009	-.0167 ± .0548	-.0118 ± .0494
Anacystis nidulans						
Mid-log inoc. BG-I	-.1145 ± .0260	-.1172 ± .0239	-.0796 ± .0137	-.0899 ± .0161	-.0614 ± .0158	-.0480 ± .0099
Contaminant redn. BG-I	-.0051 ± .0275	-.0078 ± .0099	-.0114 ± .0105	-.0177 ± .0453	-.0589 ± .1216	-.0147 ± .0059
Day-O inoc. BG-II	-.0640 ± .0320	-.0448 ± .0225	-.0596 ± .0149	-.0522 ± .0470	-.0474 ± .0136	-.0437 ± .0306
Contaminant redn. BG-II	-.0197 ± .0074	-.0213 ± .0114	-.0161 ± .0168	-.0279 ± .0140	-.0068 ± .0263	-.0323 ± .0133
Filtrate BG-VII	-.0375 ± .0684	-.0469 ± .0405	-.0212 ± .0660	-.0525 ± .1060	-.0316 ± .0873	-.0301 ± .1084
Gloeocapsa alpicola						
Mid-log inoc. BG-I	-.0688 ± .0212	-.1356 ± .0357	-.0849 ± .0246	-.1205 ± .0980	-.0484 ± .0157	-.0466 ± .0301
Contaminant redn. BG-I	-.0194 ± .0159	-.0606 ± .0075	-.0802 ± .0516	-.0462 ± .0217	-.0484 ± .0157	-.0466 ± .0301
Day-O inoc. BG-II	-.0868 ± .0328	-.0838 ± .0278	-.0494 ± .0176	-.0794 ± .0438	-.0781 ± .0272	-.0596 ± .0179
Contaminant redn. BG-II	-.0110 ± .0130	-.0198 ± .0117	-.0271 ± .0265	-.0139 ± .0104	-.0141 ± .0130	-.0213 ± .0111
Filtrate BG-VII	-.0395 ± .0437	-.0596 ± .0118	-.0656 ± .0082	-.0473 ± .0309	-.0197 ± .0460	-.0208 ± .2174
Oscillatoria chalybia						
Mid-log inoc. BG-I	-.0966 ± .0386	-.0637 ± .0518	-.1255 ± .0478	-.1153 ± .0199	-.1149 ± .0288	-.0607 ± .0108
Contaminant redn. BG-I	-.0704 ± .0311	-.0341 ± .0121	-.0393 ± .1598	-.0468 ± .0353	-.0234 ± .0272	-.0285 ± .0226
Day-O inoc. BG-II	-.0761 ± .0327	-.0571 ± .0154	-.1986 ± .0631	-.1074 ± .0450	-.0614 ± .0181	-.0905 ± .0272
Contaminant redn. BG-II	-.0110 ± .0052	-.0138 ± .0066	-.0143 ± .0087	-.0037 ± .0165	-.0006 ± .0112	-.0047 ± .0130
Filtrate BG-VII	-.0996 ± .1473	-.0963 ± .1387	-.1012 ± .2162	-.0949 ± .1623	-.0847 ± .0721	-.0805 ± .2214

Table 3-1 Continued

Series	Alcaligenes faecalis*	Enterobacter aerogenes*	Escherichia coli*	Proteus vulgaris*	Pseudomonas aeruginosa*	Serratia marcescens*
Oscillatoria formosa						
Mid-log inoc. BG-I	-.1546 ± .0224	-.0910 ± .0450	-.0864 ± .0393	-.0964 ± .0507	-.0795 ± .0379	-.0679 ± .0159
Contaminant redn. BG-I	-.0410 ± .0139	-.0278 ± .0191	-.0182 ± .0194	-.0250 ± .0272	-.0428 ± .0219	-.0451 ± .0309
Day-O inoc. BG-II	-.2957 ± .2187	-.2218 ± .5164	-.2275 ± .0640	-.2231 ± .1002	-.0751 ± .0260	-.0709 ± .0165
Contaminant redn. BG-II	-.0018 ± .0139	-.0051 ± .0158	-.0120 ± .0132	-.0124 ± .0044	-.0058 ± .0101	-.0129 ± .0352
Filtrate BG-VII	-.0761 ± .3122	-.0732 ± .1638	.0136 ± .0965	-.0678 ± .1897	-.0568 ± .0728	-.0671 ± .2056
Phormidium faveolarum						
Mid-log inoc. BG-I	-.0880 ± .0431	-.0640 ± .0460	-.0966 ± .0599	-.1782 ± .0757	-.1378 ± .0348	-.0999 ± .1055
Contaminant redn. BG-I	.0029 ± .0142	-.0368 ± .0216	-.0196 ± .0137	-.0266 ± .0347	-.0188 ± .0084	-.0515 ± .0308
Dau-O inoc. BG-II	-.0880 ± .0275	-.0594 ± .0367	-.0552 ± .0313	-.1567 ± .1194	-.2437 ± .0778	-.0456 ± .0254
Contaminant redn. BG-II	-.0195 ± .0630	-.0174 ± .0189	-.0262 ± .0235	-.0142 ± .0247	-.0047 ± .0085	-.0542 ± .2044
Filtrate BG-VII	-.0697 ± .1664	-.0841 ± .1405	-.0646 ± .2584	-.0643 ± .0767	-.0704 ± .1549	-.0811 ± .1332
Ankistrodesmus braunii						
Mid-log inoc. G-I	-.0701 ± .0193	-.0764 ± .0172	-.0365 ± .0290	-.0756 ± .0288	-.1129 ± .0504	-.0620 ± .0438
Day-O inoc. G-II	-.0857 ± .0435	-.0801 ± .1205	-.0542 ± .1598	-.0745 ± .3424	-.1977 ± .0297	-.1753 ± .0254
Filtrate G-VII	-.0769 ± .2780	-.0965 ± .1696	-.0520 ± .0090	-.0904 ± .0590	-.0890 ± .0751	-.0558 ± .1196
Chlorella pyrenoidosa						
Mid-log inoc. G-I	-.1586 ± .0219	-.1013 ± .0781	-.0624 ± .0305	-.1800 ± .1094	-.0985 ± .0767	-.0855 ± .0481
Day-O inoc. G-II	-.0743 ± .0942	-.1265 ± .0148	-.0763 ± .1384	-.0552 ± .0419	-.0574 ± .1100	-.0552 ± .0787
Filtrate G-VII	-.0925 ± .2509	-.0851 ± .2041	-.0708 ± .3320	-.0830 ± .2321	-.0550 ± .3776	-.0686 ± .3630
Chlorella vulgaris						
Mid-log inoc. G-I	-.1255 ± .1253	-.0949 ± .0858	-.0464 ± .0480	-.1826 ± .4619	-.1092 ± .0581	-.0651 ± .0547
Day-O inoc. G-II	-.1003 ± .0137	-.1138 ± .0155	-.0407 ± .1286	-.1352 ± .0176	-.1901 ± .0171	-.0989 ± .0148
Filtrate G-VII	-.0852 ± .1884	-.0656 ± .1201	-.0553 ± .1392	-.0757 ± .1238	-.0683 ± .2253	-.0644 ± .2491

Table 3-1 Continued

Series	*Alcaligenes faecalis**	*Enterobacter aerogenes**	*Escherichia coli**	*Proteus vulgaris**	*Pseudomonas aeruginosa**	*Serratia marcescens**
Scenedesmus obliquus						
Mid-log inoc. G-I	-.0458 ± .0365	-.0574 ± .0238	-.0583 ± .0215	-.0891 ± .0506	-.0777 ± .0357	-.0974 ± .0354
Day-O inoc. G-II	-.0655 ± .0142	-.0792 ± .0065	-.0541 ± .0631	-.0629 ± .0093	-.0466 ± .2023	-.0492 ± .1126
Filtrate G-VII	-.0324 ± .0214	-.0508 ± .0307	-.0401 ± .0352	-.0151 ± .0809	-.0550 ± .0852	-.0325 ± .0908
Mixed blue-greens, single bacteria inoc. BG-VIII	-.2463 ± .3895	-.2368 ± .5988	-.1666 ± .1590	-.2392 ± .6491	-.1730 ± .1538	-.1479 ± .2096
Mixed blue-greens, mixed bacteria inoc. BG-IX	-.2600 ± .4379	-.2741 ± .7115	-.1081 ± 2.433	-.1632 ± .4947	-.1536 ± .1237	-.1397 ± .0671
Mixed greens, single bacteria inoc. G-VIII	-.1635 ± .0689	-.1462 ± .0353	-.1280 ± .0773	-.1912 ± .4282	-.1744 ± .0601	-.1493 ± .0567
Mixed greens, mixed bacteria inoc. G-IX	-.1176 ± .3440	-.2082 ± .2771	-.1417 ± .1001	-.1743 ± .5893	-.1493 ± .1280	-.1579 ± .0885
Brevibacterium sp., effect on dieoff of ...	-.0513 ± .0300	-.0494 ± .0127	-.0755 ± .0123	-.0951 ± .0178	-.1011 ± .0407	-.0624 ± .0168
Flaveobacterium sp., effect on dieoff of ...	-.1437 ± .0945	-.1616 ± .0743	-.0947 ± .0431	-.1520 ± .0306	-.1042 ± .0519	-.0666 ± .0333
Bacteria alone, dieoff in algal growth medium, VI	-.0228 ± .0279	-.0097 ± .0317	-.0214 ± .0263	-.0149 ± .0325	-.0133 ± .0222	-.0098 ± .0283
Bacteria alone, anaerobic dieoff rates of ...	-.0131 ±. 0253	-.0352 ± .0175	-.0490 ± .0072	-.0131 ± .0307	-.0315 ± .0259	-.0563 ± .0108

Table 3-1 Continued

Series	*Alcaligenes faecalis**	*Enterobacter aerogenes**	*Escherichia coli**	*Proteus vulgaris**	*Pseudomonas aeruginosa**	*Serratia marcescens**
Growth rates during Series I						
Anabaena cylindrica						
Control .0130 ± .0021	.0117 ± .0099	.0107 ± .0079	.0043 ± .0139	.0114 ± .0143	.0155 ± .0101	.0152 ± .0093
Anacystis nidulans						
Control .0171 ± .0111	.0116 ± .0148	.0112 ± .0101	.0131 ± .0034	.0064 ± .0083	.0102 ± .0173	.0113 ± .0095
Gloeocapsa alpicola						
Control .0041 ± .0071	.0083 ± .0128	.0070 ± .0072	.0085 ± .0076	.0039 ± .0109	.0049 ± .0050	.0119 ± .0060
Oscillatoria chalybia						
Control .0121 ± .0021	.0084 ± .0059	.0067 ± .0048	.0049 ± .0123	.0189 ± .0078	.0155 ± .0101	.0152 ± .0093
Oscillatoria formosa						
Control .0290 ± .0077	.0285 ± .0174	.0256 ± .0115	.0209 ± .0146	.0160 ± .0197	.0101 ± .0176	.0159 ± .0187
Phormidium faveolarum						
Control .0025 ± .0018	.0013 ± .0116	.0005 ± .0094	.0027 ± .0088	.0048 ± .0148	.0019 ± .0118	.0059 ± .0119
Ankistrodesmus braunii						
Control .0085 ± .0021	.0074 ± .0051	.0042 ± .0061	.0087 ± .0071	.0027 ± .0031	.0049 ± .0008	.0111 ± .0029
Chlorella pyrenoidosa						
Control .0199 ± .0121	.0143 ± .0091	.0171 ± .0061	.0171 ± .0091	.0199 ± .0210	.0100 ± .0067	.0121 ± .0077
Chlorella vulgaris						
Control .0102 ± .0037	.0177 ± .0041	.0120 ± .0141	.0200 ± .0040	.0099 ± .0061	.0154 ± .0071	.0136 ± .0088
Scenedesmus obliquus						
Control .0075 ± .0065	.0071 ± .0091	.0089 ± .0072	.0101 ± .0214	.0116 ± .0100	.0171 ± .0065	.0144 ± .0061

*$\log_{10}\text{day}^{-1}$

Additional data on dieoff effects of enteric bacteria were obtained using a typical soil inhabitant blue-green algal species, *Nostoc muscorum*. Filtrate from an actively growing axenic culture of this organism was exposed to the enteric bacteria in the same manner as the other filtrate series. Since *Nostoc* is not ordinarily found as a phytoplankton member in waste stabilization ponds, all of the series of combinations were not run on this organism. The data for the filtrate run with *Nostoc* (Appendix A, Table A-50) are included and intended for comparison with the data for the other enteric bacterial-algal runs. The coefficients were: *Alcaligenes*, -.0862 ± .2171 day^{-1}; *Enterobacter*, -.1021 ± .1180 day^{-1}; *Escherichia*, -.0881 ± .1177 day^{-1}; *Proteus*, -.1021 ± .0078 day^{-1}; *Pseudomonas*, -.0639 ± .1352 day^{-1}; *Serratia*, -.6020 ± .0953 day^{-1}. These are comparatively higher rates than those obtained for many of the filtrate runs involving the other six species of blue-greens. The most pronounced effect on any bacteria by *Nostoc* was that exerted on *Serratia*. This bacteria appeared to be more resistant as compared to the other species, yet the coefficient was -0.602 ± .0953 day^{-1}.

Few genera were found to be persistent contaminants of the blue-green algal cultures. *Brevibacterium* and *Flaveobacterium* were the two most frequent contaminants, occurring primarily in filamentous blue-green species. Coefficients for these two bacterial genera in control runs of the test blue-green algae are shown in Table A-25, Appendix A. When enteric bacteria were present, definite inhibitory effects were noted in the coefficients for the culture contaminants (Table 3-1). At the same time, pronounced antagonistic effects of the enteric bacteria were noted, exerted by the contaminants. Coefficients were higher when the enterics were in the presence of *Flaveobacterium* as compared to *Brevibacterium*. Coefficients for enteric dieoffs were higher when in the presence of *Flaveobacterium* alone than when in the presence of many of the axenic algal cultures, further evidence that blue-green protoplasmic constituents were furnishing nutritional compounds to the enteric bacteria.

A comparison of these coefficients with those obtained when the enteric bacteria were placed in the sterile algal growth medium (Series VI control) is noteworthy because of the differences which occurred as a result of any biological antagonism. Dieoff of enteric bacteria in algal growth medium under anaerobic and aerobic conditions was similar, for the most part, to dieoff when the enterics were present with the algae (Series BG-I, BG-II). Considering the trace quantities of nutritive organics which were present in the algal growth medium, those data demonstrate the persistent nature of the enteric bacteria and their ability to survive in situations which would be considered inadequate for life support of the bacteria.

Of the four species of green algae studied, Chlorella pyrenoidosa and C. vulgaris exerted more antagonism than did Scenedesmus obliquus or Ankistrodesmus braunii. Approximately similar dieoff rates for the enteric bacteria occurred when in the presence of Scenedesmus obliquus and Ankistrodesmus braunii and all the blue-green algae tested. Possibly Chlorella produced some substance such as chlorellin which was responsible for the accelerated dieoff of the enterics. Numerically larger coefficients were obtained for the series employing filtrate from the green algae (G-VII). Most of these coefficients were higher than those developed by the blue-green algae under similar circumstances.

Perhaps the most significant runs were those incorporating mixed algae and the additions of single bacterial and mixed bacterial species. The resulting coefficients are shown in Table 3-1. Competition among algal species for survival apparently accelerated the dieoff of the bacteria. Coefficients calculated for the individual bacterial species were similar to the rates for the same individual bacterial species when in mixed culture (comparing BG-VIII and BG-IX; G-VIII and G-IX). Oddly enough, in these series a significant number of the coefficients were higher for the mixed blue-green species than for the mixed green species. These data infer that the blue-green algae secreted antibacterial substances when in the presence of other blue-greens, whereas the green algae tested secreted their antibacterial materials in heterogeneous populations or in axenic culture. No runs were conducted with green algal contaminants due to their near total absence from the cultures of green algae. For the mixed enterics with mixed blue-green algae and separately with mixed green algae, coefficients were computed for the total numbers of enterics present. The data are as follows: mixed blue-greens and enterics, -0.1536 ± .0990 day^{-1}; mixed green algal species and enterics, -.1487 ± .0935 day^{-1}. These coefficients would correspond to what is ordinarily considered to be a "total coliform" count dieoff coefficient.

Only a small but significant part of the total research effort was devoted to establishing the effects of the presence of enteric bacteria with algae. Biomass of controls (axenic algal cultures) were compared with samples taken during Series I runs. Coefficients representing these effects are presented last in Table 3-1. In some instances, the presence of the enteric bacteria effected a reduction in the total biomass productivity of the test algal species.

Pathogenic Bacteria Dieoff Studies

Five species of pathogenic bacteria were subjected to tests which were similar to those involving the enteric bacteria. The dieoff coefficients for those series are presented in Table 3-2 and Tables A-58 through A-72, Appendix A. Considering the difficulty encountered in maintaining those pure cultures of pathogens in the laboratory, their dieoff was slower when in the presence of algae; however, no aftergrowth was found for any of those bacterial species. Considering the coefficients in Table 3-2, it would appear that the blue-green and green algal test species had approximately the same effect on those bacteria as they did on the enteric species. Surprisingly, the mixed algal cultures did not exert as great an effect on the dieoff coefficients of the pathogens as on those of the enteric bacteria. Coefficients produced under anaerobic conditions were significantly lower than the rates in the same medium under aerobic conditions. Therefore it may be concluded that the algae had little effect on the pathogenic bacterial species.

Aftergrowth Potential Measurements

Extending the duration of the runs involving the test bacteria permitted evaluation of one of the original purposes for this investigation; namely, identification of any aftergrowth potentials of each bacterial species tested. Of the eleven species of bacteria tested, three demonstrated abilities to regenerate their populations. These were *Serratia marcescens*, *Pseudomonas aeruginosa*, and less frequently, *Escherichia coli*. The other enteric bacterial species as well as the pathogens apparently did not possess this

Table 3-2. Dieoff Coefficients for Series Utilizing Pathogenic Bacterial Species.

Series	Salmonella paratyphi*	Salmonella typhosa*	Shigella paradysenteriae*	Shigella dysenteriae*	Vibrio comma*
Mid-log inoculation of ...					
Anabaena cylindrica	-.0751 ± .0098	-.0601 ± .0351	-.0707 ± .0141	-.0742 ± .0139	-.0511 ± .0137
Anacystis nidulans	-.0840 ± .0174	-.0758 ± .0157	-.0981 ± .0317	-.1249 ± .0208	-.0997 ± .0247
Gloeocapsa alpicola	-.0657 ± .0401	-.0609 ± .0156	-.0832 ± .0087	-.0745 ± .0091	-.0755 ± .0329
Oscillatoria chalybia	-.0613 ± .0309	-.0523 ± .0303	-.0673 ± .0330	-.0717 ± .0178	-.0639 ± .0082
Oscillatoria formosa	-.0839 ± .0199	-.0684 ± .0093	-.0622 ± .0071	-.0688 ± .0217	-.0706 ± .0149
Phormidium faveolarum	-.0790 ± .0106	-.0602 ± .0195	-.0791 ± .0181	-.0670 ± .0186	-.0658 ± .0166
Ankistrodesmus braunii	-.0726 ± .0193	-.0658 ± .0181	-.0730 ± .0250	-.0707 ± .0272	-.0593 ± .0146
Chlorella pyrenoidosa	-.0950 ± .0495	-.0986 ± .0386	-.0996 ± .0261	-.0840 ± .0520	-.0747 ± .0383
Chlorella vulgaris	-.0669 ± .0334	-.0572 ± .0317	-.0633 ± .0322	-.0700 ± .0261	-.0465 ± .0283
Scenedesmus obliquus	-.0833 ± .0276	-.0700 ± .0381	-.1061 ± .0411	-.0935 ± .0440	-.0872 ± .0480
Mixed blue-green species	-.0759 ± .0259	-.0775 ± .0244	-.0564 ± .0187	-.1124 ± .0673	-.0933 ± .0272
Mixed green species	-.1553 ± .0617	-.1156 ± .0402	-.1702 ± .0586	-.1345 ± .0457	-.1460 ± .0535
Dieoff rates in algal growth medium, Aerobic	-.0622 ± .0454	-.0775 ± .0174	-.0738 ± .0276	-.0728 ± .0256	-.0625 ± .0204
Dieoff rates in algal growth medium, Anaerobic	-.0207 ± .0293	-.0268 ± .0262	-.0492 ± .0146	-.0194 ± .0346	-.0162 ± .0211

*log$_{10}$day^{-1}

capability under the conditions of testing during these experiments. Data for the aftergrowth, and times of occurrences in the test periods are presented in Table 3-3. Aftergrowth was caused by the readily available protoplasmic constituents of the algae as the algae reached their declining or log death phase. The danger of recurrence of these bacterial species which showed the aftergrowth potential is therefore present when sufficient organic nutrients are present in the surrounding aquatic environment. And further, regardless of the efficiencies of removal of any treatment process or design parameter such as waste stabilization ponds, if absolutely 100 percent kill of these bacteria is not accomplished, aftergrowth can indeed occur in the effluent receiving-waters.

Organic Carbon Production by Algae

Little is known concerning the contribution by algae to the organic carbon content of waters and the resulting effects of the organic carbon on such parameters as bacterial survival or reproduction capacities. During Series BG-I, G-I, BG-VIII, and G-VIII, measurements were made at the 90-day time period in an attempt to determine the maximum yield of total carbon and total organic carbon by the algae, or biomass present in culture. These data are presented in Table 3-4. Significant amounts of organic carbon were present in the cultures after 90-days of testing. Comparison with the controls reveals by yet another method that some inhibition by the enteric bacteria on the overall productivity of the algae occurred. The contribution by the bacteria to the organic carbon content was negligible in all cases. This can be proven due to the fact that, on the average, it takes 10^{12} bacterial cells to equal one milligram of biomass weight and the cells are obviously not totally organic carbon. Consequently, the total organic carbon values, as presented in Table 3-4, may be assumed to have been derived from the algae themselves. These levels of organic carbon represent adequate quantities for, at least, the survival of the enteric bacteria, if not multiplication of same over a period of time.

Table 3-3. Aftergrowth Characteristics of Enteric Bacterial Species with Single Species of Algae (Series I).

Algal Species	Bacterial Genera	Min. No. Bacteria in run, No/ml	Day Min. No. Occurred	Aftergrowth, Max. No/ml	Day Max. No. Occurred
Anabaena cylindrica	Pseudomonas	10,000	63	180,000	70
	Serratia	6,000	63	800,000	84
Anacystis nidulans	Pseudomonas	33,000	63	310,000	70
	Serratia	48,000	63	340,000	84
Gloeocapsa alpicola	Serratia	250	56	8,110	91
Oscillatoria chalybia	Pseudomonas	310	56	160,000	91
Phormidium faveolarum	Escherichia	<100	56	1,920	84
	Pseudomonas	<100	63	3,300	70
	Serratia	<100	42	3,320	56
Ankistrodesmus braunii	Pseudomonas	<2,000	56	280,000	91
	Serratia	50,400	63	542,000	91
Chlorella pyrenoidosa	Pseudomonas	1,030	77	525,000	91
	Serratia	286	77	38,100	91
Chlorella vulgaris	Pseudomonas	132	63	4,000	91
Scenedesmus obliquus	Escherichia	324	63	64,200	91
	Pseudomonas	4,600	63	60,000	91
	Serratia	1,290	63	41,000	91

Table 3-4. Total Carbon and Total Organic Carbon Content of Biomass After Ninety Days (Series BG-I, G-I, BG-VIII, and G-VIII, in mg/l).

Algal Species	Bacteria Added to Algal Culture: Alcaligenes faecalis		Enterobacter aerogenes		Escherichia coli		Proteus vulgaris		Pseudomonas aeruginosa		Serratia marcescens		Control	
	T.C.	T.O.C.	T.C.	T.O.C.	T.C.	T.O.C.	T.C.	T.O.C.	T.C.	T.O.C.	T.C.	T.O.C.	T.C.	T.O.C.
Anabaena cylindrica	47	41	60	58	47	33	53	41	51	41	49	41	77	47
Anacystis nidulans	36	34	49	45	36	34	38	36	33	33	41	35	52	47
Gloeocapsa alpicola	64	56	67	58	71	66	77	62	64	60	58	53	67	67
Oscillatoria chalybia	128	124	79	69	64	60	56	56	62	62	86	75	65	42
Oscillatoria formosa	100	88	198	154	206	166	200	166	252	198	120	75	172	168
Phormidium faveolarum	77	73	81	73	69	69	104	77	73	71	69	69	85	82
Ankistrodesmus braunii	62	41	57	43	67	52	47	40	47	38	59	48	54	50
Chlorella pyrenoidosa	50	37	61	50	47	40	51	45	56	47	56	47	97	96
Chlorella vulgaris	53	41	59	43	60	51	54	50	58	49	55	39	54	33
Scenedesmus obliquus	60	41	62	40	61	50	51	41	53	38	57	55	51	39
Mixed Blue-greens BG-VIII	92	58	83	49	118	95	169	124	95	67	77	58		
Mixed Greens G-VIII	65	61	67	65	137	63	61	47	130	81	94	75		

CHAPTER 4

LABORATORY AND FIELD WASTE STABILIZATION POND STUDIES

The objective of these experiments were to establish dieoff coefficients for selected species of bacteria under conditions which would occur in operational waste stabilization ponds. Two kinds of pond systems were investigated; namely, laboratory scale units which were designed on a volume detention time basis to closely correspond to the field units, and field scale pilot units.

Laboratory Waste Stabilization Pond Studies

A diagram of the laboratory waste stabilization ponds depicting the three different design concepts is shown in Figure 4-1. Throughout the test period of approximately 60 days, supplementary data were taken on phytoplankton populations to relate their concentrations to possible effects on the bacterial populations. The procedure was as follows. A liter of domestic wastewater was added to each of Series I, II, and III daily. Series I differed from Series II and III in that Series I began the treatment cycle with a six-liter anaerobic pretreatment chamber. Also, on a daily basis, 0.5 liter of effluent from the anaerobic unit was added to each of the two following facultative units. This was followed by the addition of one liter, combined from each of the two facultative units, to the maturation pond. In Series I the volume of the facultative units was 90 liters and that of the maturation pond was 18 liters. These volumes provided detention times of 6, 90, and 18 days respectively. The maturation ponds in Series I, II, and III were similar.

Series II facultative units provided anaerobic treatment in an anaerobic trench located at the influent end of each unit. The volume of these facultative units was 84 liters. The facultative units and the maturation pond unit in Series III had the same volume as each of the similar units in Series I. In Figure 4-1 the locations of bacterial inoculation points (i) and sampling stations (numbers) are shown. Duplicated sampling station

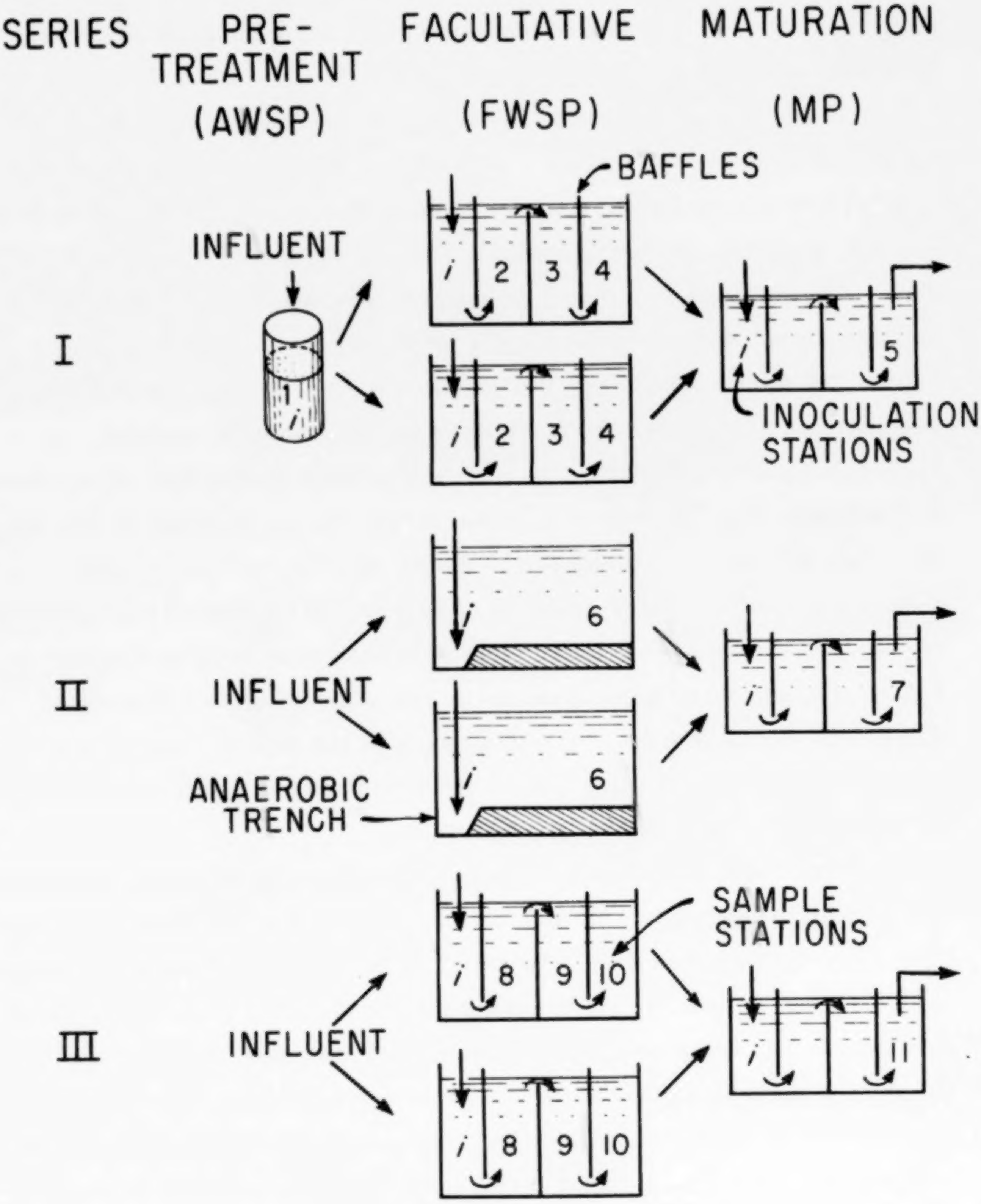
SERIES
PRE-TREATMENT (AWSP)
FACULTATIVE (FWSP)
MATURATION (MP)
BAFFLES
INFLUENT
I
1
2
3
4
5
INOCULATION STATIONS
II
INFLUENT
6
7
ANAEROBIC TRENCH
III
INFLUENT
SAMPLE STATIONS
8
9
10
11

FIG. 4-1. SCHEMATIC OF LABORATORY PONDS

numbers mean that equivalent volumes were sampled at those points and mixed prior to analysis. Overall, the laboratory waste stabilization pond units proved to be amenable to bacteriological analysis because of their relatively small size, which permitted accurate bacterial inoculations.

Data for all bacteria counts were obtained by plating duplicates of two dilutions from each sample. These data, representing the statistical mean values of four counts per sample are presented in Appendix B as Tables B-1 through B-12. Blanks in these tables are the result of inconsistent plating; or the types or species of bacteria in question did not appear on the plates on that date; or the counts were too high to be statistically valid. The gaps do not imply the absence of the bacteria. Incubation periods were not consistent throughout the tests for the following reasons. In many instances it was found that room temperature incubation, as opposed to incubation at 35°C, enhanced some of the coliform species as well as other bacterial species found on the plates used for the total counts. Also, in several instances, periods of 72 hours of incubation were necessary to obtain representative counts. This peculiar characteristic of many bacterial species has been observed by the authors before. Therefore, these data provide a more accurate account of the actual numbers of bacteria present at the sampling times than would have been recorded by incubation for, say, only 24 hours at 35°C.

On July 7 and 29 cultures of Escherichia coli, Pseudomonas aeruginosa, and Serratia marcescens were inoculated into the selected locations in the laboratory units (Figure 4-1). The total bacterial numbers for each inoculum are presented in Table 4-1. By taking into account the daily additions of wastewater, its complement of the bacteria in question, volumes of the laboratory ponds and other pertinent quantitative physical factors, dieoff coefficients for each of the three added species were calculated. These coefficients are shown in Table 4-2 for the vicinities adjacent to the point of inoculation. In the majority of cases the dieoff coefficients were much higher than those found for the axenic culture experiments. The exceptions to this can be seen in the data in Table 4-2 as for, by way of example, Pseudomonas aeruginosa at station 2. Obviously, conditions prevailing

Table 4-1. Bacteria Inoculated Into Selected Stations In Laboratory Waste Stabilization Ponds

	Species and Date					
	Escherichia coli		*Pseudomonas aeruginosa*		*Serratia marcescens*	
Station*	July 7	July 29	July 7	July 29	July 7	July 29
1	13.2406**	9.3010	11.4700	9.9030	9.9607	9.1760
2	12.2579	13.8000	12.1847	11.6020	11.9240	11.3010
5	12.4595	9.0000	12.3180	11.6535	11.1760	9.5440
6	11.6730	11.3656	11.7136	10.0608	12.0415	--
7	13.1105	9.3980	11.8720	11.9030	12.6445	--
8	12.5841	--	11.6200	11.4775	13.4345	9.6020
11	11.2480	--	11.7780	9.7401	10.2600	11.0000

*Locations indicated in Figure 4-1.

**$\log_{10}$ total number inoculated.

Table 4-2. Dieoff Coefficients of Inoculated Bacteria in Laboratory Scale Waste Stabilization Ponds

	Coefficients for Bacterial Species (day^{-1})		
Station Inoculated*	*Escherichia coli*	*Pseudomonas aeruginosa*	*Serratia marcescens*
1	-1.03	-0.34	-0.43
2	-0.44	-0.14	-0.41
5	-0.37	-0.93	-1.38
6	-0.31	-1.00	-1.78
7	-0.61	-1.10	-1.53
8	-0.37	-0.63	-1.16
11	-0.51	-1.26	-1.15

*Inoculation point at or adjacent to indicated station; Figure 4-1.

in the early stages of the treatment units caused higher dieoff coefficients to occur than did those in the later stages. Anaerobic pretreatment did not cause the accelerated dieoff for the three inocula bacteria species, expecially Pseudomonas and Serratia, as was expected. It might be concluded that the corresponding decreases in nutrient materials aided in acceleration of that dieoff which did occur. It should be noted, however, that the additions of the inocula bacteria at station 8 resulted in reduced dieoff coefficients for two of the three test species. Fewer algae were present at that station during part of the test period than were present at other stations except station 11 (Table 4-4).

The reduced phytoplanktonic concentrations throughout all runs involving the laboratory ponds in Series III appeared to be a characteristic of that series throughout the test period. Of the three test bacteria, E. coli appeared to possess the greatest capacity for survival through all three types of pond combinations. Many of the enteric bacteria remained in the final effluents. Examination of all three effluents revealed that the quality of the effluent permitted some aftergrowth of all three test bacteria species. The increases in numbers of bacteria did not exceed two orders of magnitude. However, the mere fact that aftergrowth did occur is in itself additional evidence that a much greater understanding of the behavior of these bacteria in ponds is needed.

Relationships between the bacteriological concentrations and the corresponding phytoplankton concentrations may be observed by referring to Tables 4-3 and 4-4 and Figures 4-2 through 4-5. Surprisingly few diatoms were present in any of the treatment units during the course of the study. Euglena sp. did not appear until on or slightly prior to August 20. For the purposes of this investigation the intermittent appearances of representative species of these divisions (Euglenophyta and Chrysophyta) permitted evaluation of the two divisions which were of primary concern, the blue-green algae (Cyanophyta) and the green algae (Chlorophyta).

Considering the behavior of the test species in the laboratory ponds, the following observations were made during the duration of the experiments. Periodic increases in concentrations of total bacteria, as noted for day 36

Table 4-3. Phytoplankton Found in Laboratory Waste Stabilization Ponds.

Algal Division	Date and Station Number								
	July 9			July 23			August 6		
Station:	3	6	9	3	6	9	3	6	9
Cyanophyta	30,000* (23,000)**	37,000 (36,000)	-0-	17,000 (31,500)	52,000 (47,000)	4,000 (7,500)	10,700 (7,000)	37,200 (60,000)	-0-
Euglenophyta	-0-	-0-	-0-	-0-	-0-	-0-	-0-	-0-	-0-
Chrysophyta	1,000 (1,000)	-0-	-0-	-0-	1,000 (1,000)	-0-	-0-	-0-	-0-
Chlorophyta	1,400 (17,000)	1,500 (2,000)	6,700 (8,000)	3,000 (6,000)	43,000 (187,000)	10,000 (20,000)	2,700 (28,000)	6,000 (23,000)	4,000 (36,000)
Total	32,400 (41,000)	38,500 (38,000)	6,700 (8,000)	20,000 (37,500)	96,000 (235,000)	14,000 (27,500)	13,400 (35,000)	43,200 (290,000)	4,000 (36,000)

Table 4-3 Continued

Algal Division	Date and Station Number		
	August 20		
Station:	3	6	9
Cyanophyta	17,000 (5,000)	13,000 (144,100)	4,000 (21,500)
Euglenophyta	1,000 (1,000)	500 (400)	100 (100)
Chrysophyta	-0-	-0-	-0-
Chlorophyta	10,000 (34,000)	19,500 (60,500)	6,900 (38,400)
Total	28,000 (40,000)	33,000 (205,000)	11,000 (60,000)

*Areal Standard Units of phytoplankton per ml.

**No. of phytoplankton per ml.

Table 4-4. Total Phytoplankton Concentrations Found In Laboratory Waste Stabilization Ponds

Date	Station Number					
	3	5	6	7	9	11
July 9	32,400* (41,000)**	4,100 (12,000)	38,500 (38,000)	100 (270)	6,700 (8,000)	3,000 (6,000)
July 23	20,000 (37,500)	6,100 (11,000)	96,000 (235,000)	2,500 (3,500)	14,000 (27,500)	3,750 (13,000)
Aug. 6	13,400 (35,000)	6,300 (16,000)	43,200 (290,000)	41,900 (52,500)	4,000 (36,000)	1,670 (4,800)
Aug. 20	28,000 (40,000)	13,000 (27,500)	33,000 (205,000)	20,000 (31,500)	11,000 (60,000)	9,600 (27,000)

*Areal Standard Units of phytoplankton per ml.
**( ) No. phytoplankton per ml.

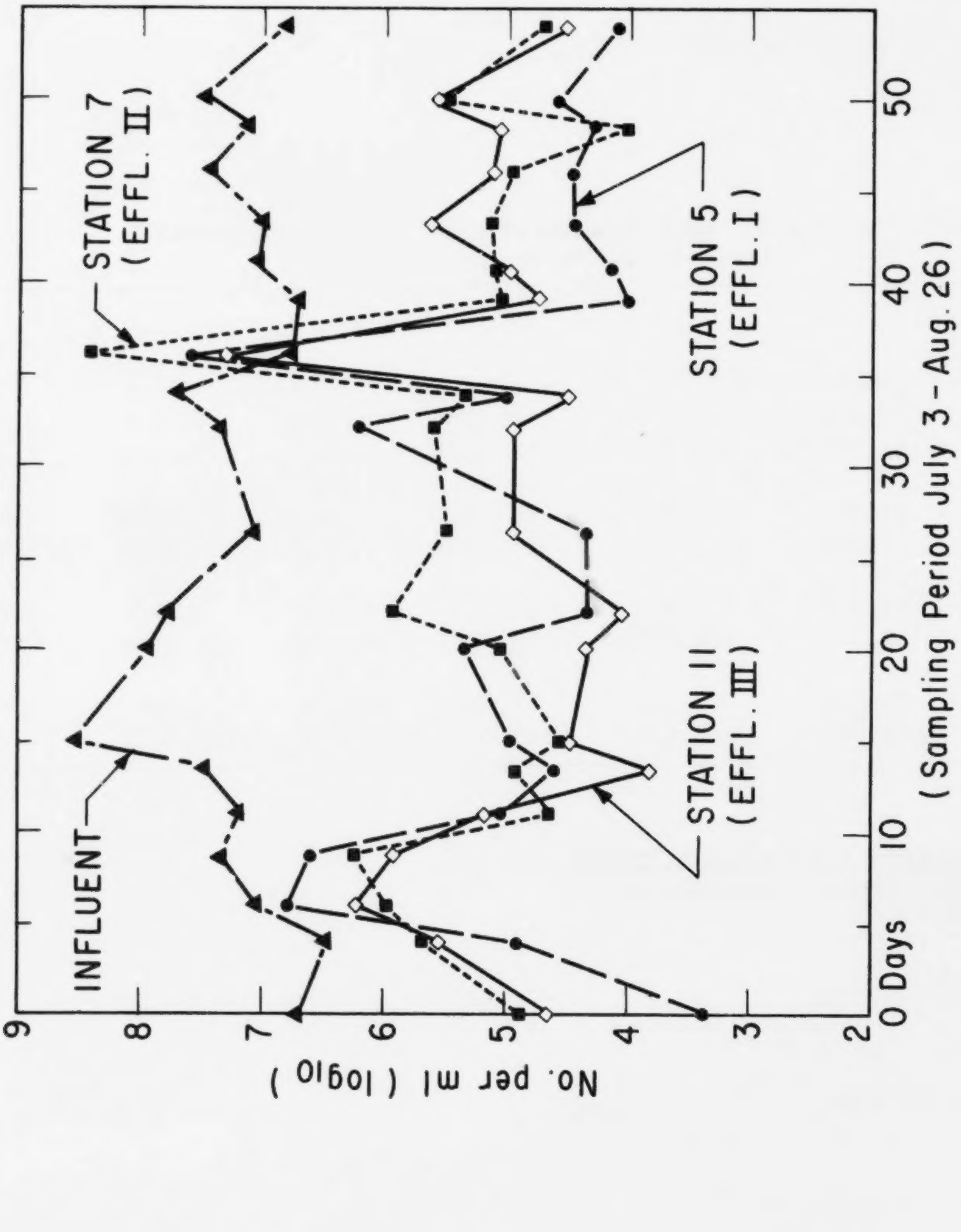

FIG. 4-2. TOTAL BACTERIA, LABORATORY PONDS

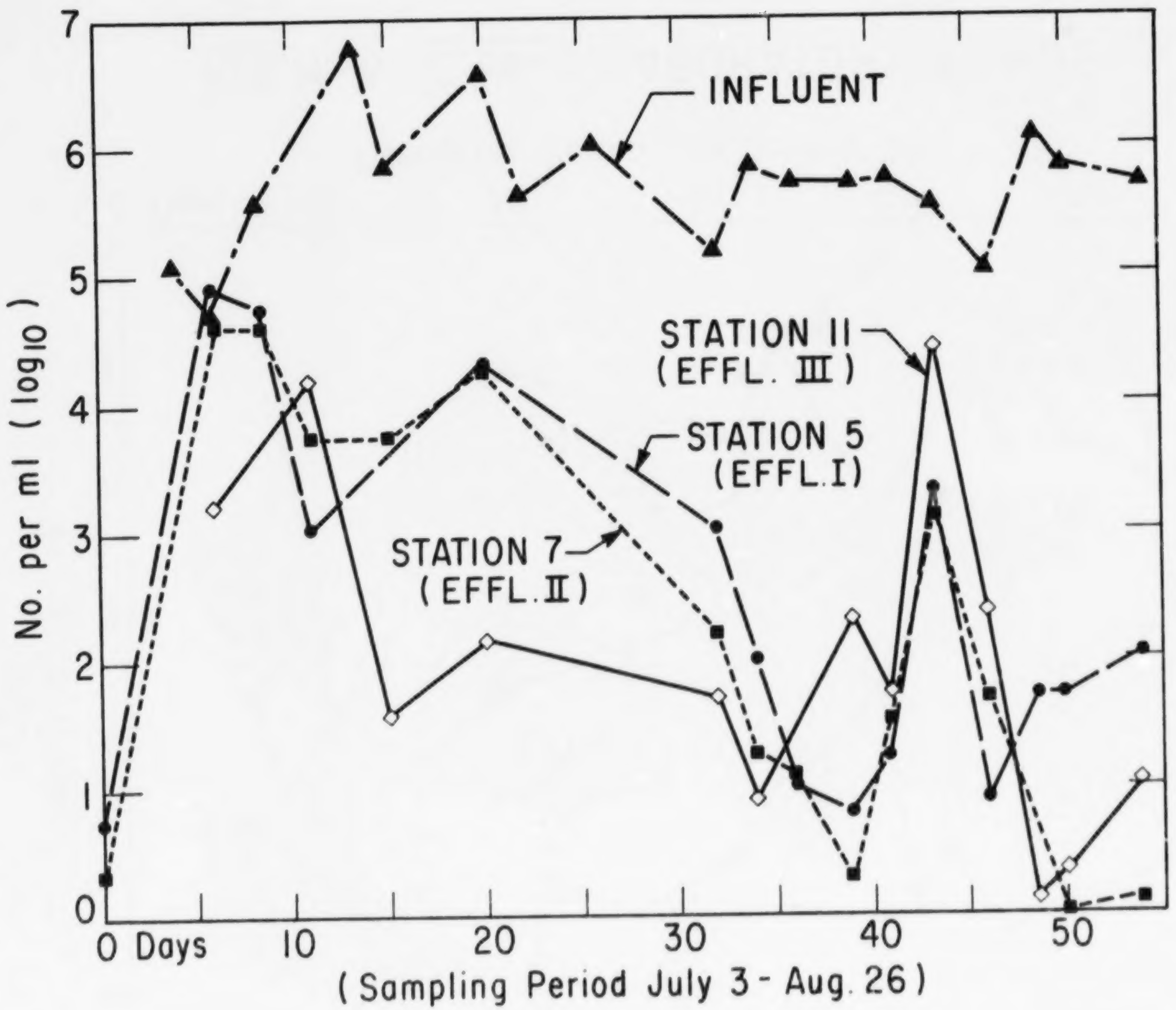

FIG. 4-3. TOTAL COLIFORM, LABORATORY PONDS

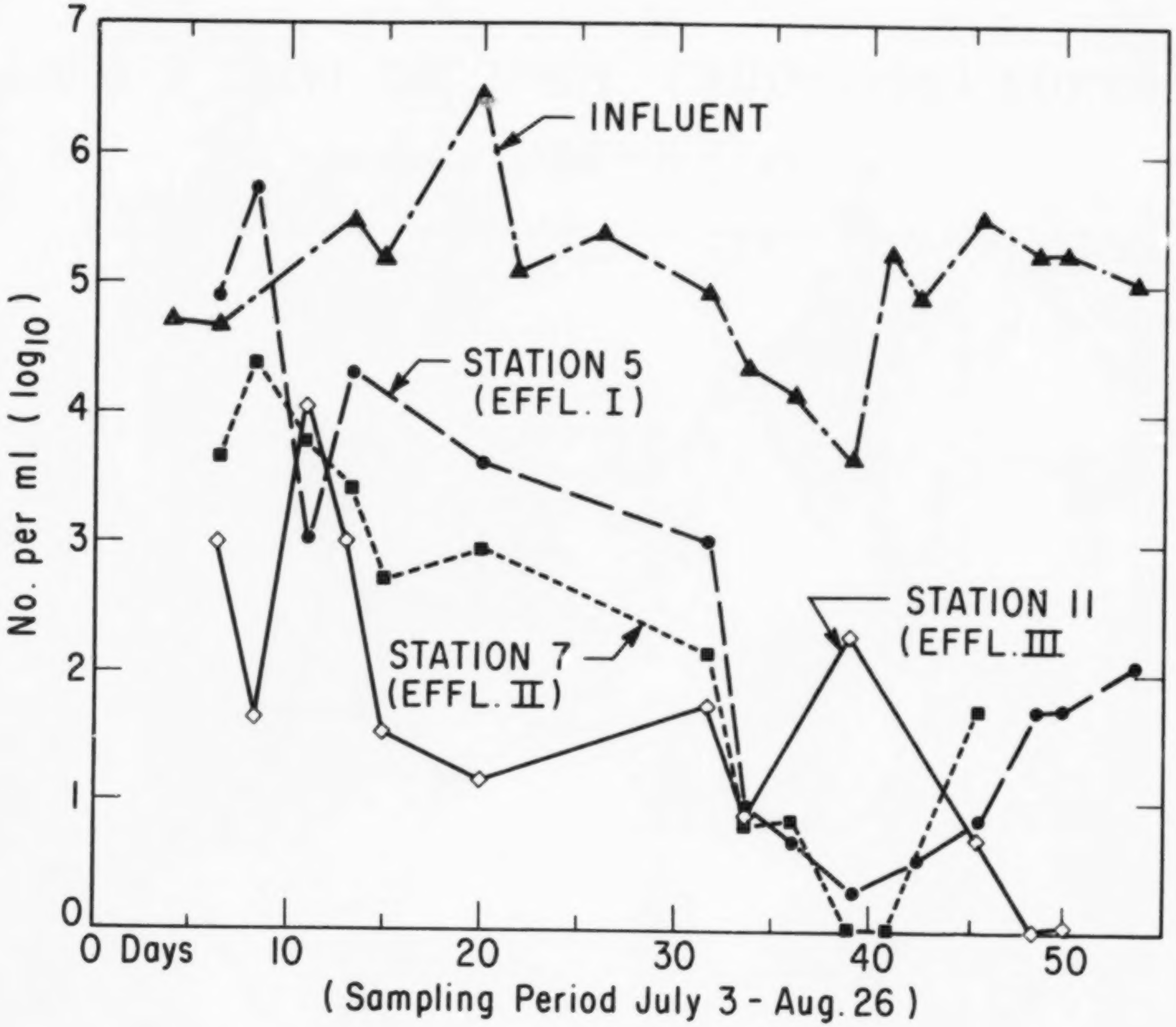

FIG. 4-4. *E. coli*, LABORATORY PONDS

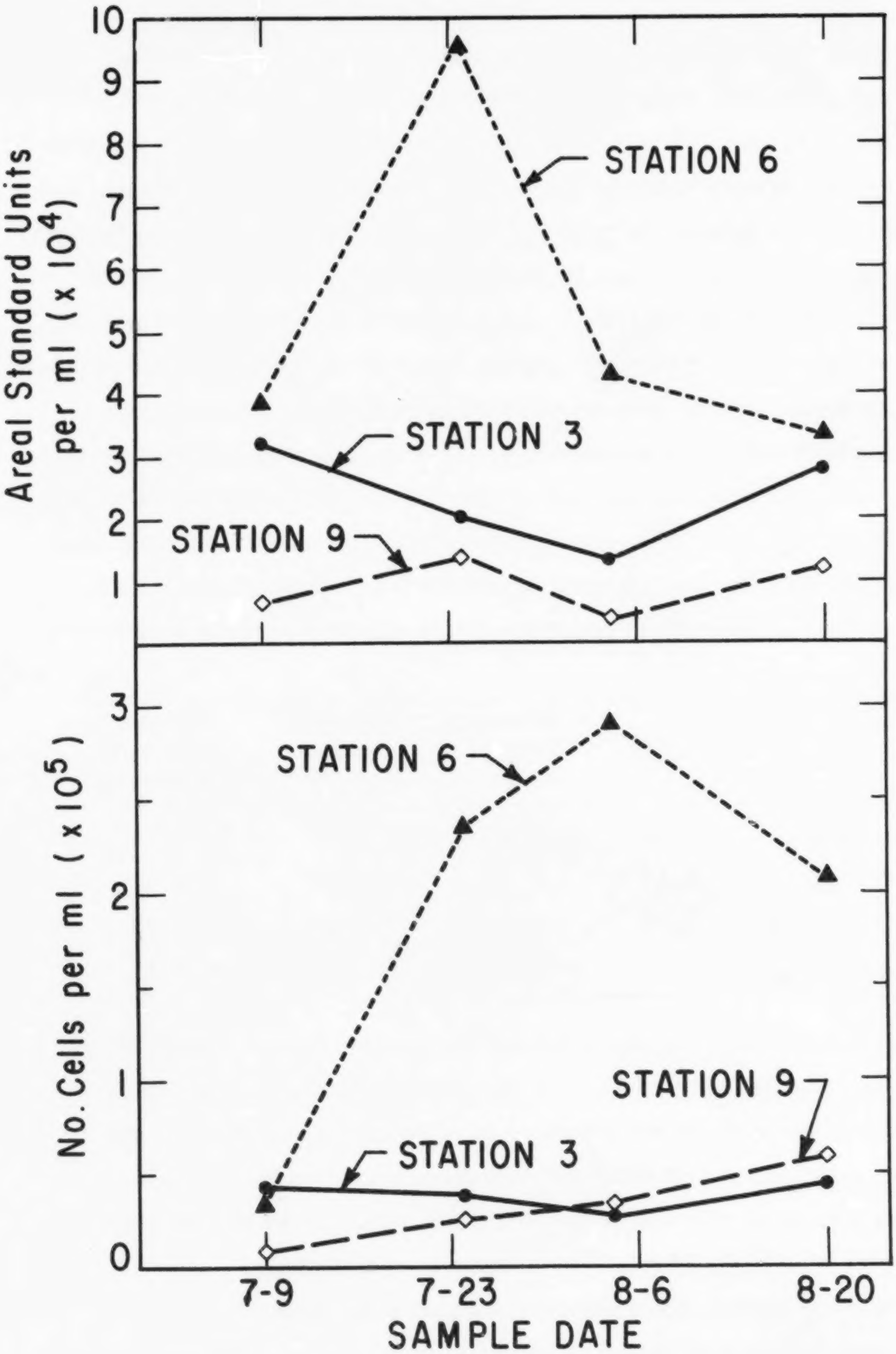

FIG. 4-5. PHYTOPLANKTON DENSITIES IN LABORATORY PONDS

(Figures 4-2), were followed closely by a sudden increase in the total coliform count (day 44, Figure 4-3). Only a small fraction of the total coliform count was attributed to E. coli, per se. Examination of the plates for the samples in question revealed colonial morphology similar to that for Enterobacter, Alcaligenes, or Proteus, rather than Pseudomonas and Serratia. The total coliform count as well as the populations of E. coli decreased significantly on or about July 23 to about August 6 when the phytoplankton populations were relatively high. During these times the blue-green population was in the majority. Figures 4-2 through 4-4 relate other pertinent points. Acclimitization of the pond systems was occurring for approximately thirty days prior to any stabilization of the effluent quality so far as total coliform or E. coli concentrations were concerned. Statistical means were calculated for all values of effluent concentrations. These indicate the systems' overall capability in reduction of the group or species in question. Data for these follow:

	Influent	Effluent Series I (Station 5)	Effluent Series II (Station 7)	Effluent Series III (Station 11)
Total Bacteria	6.92	4.95	5.37	5.13
Total Coliform	5.87	2.36	2.34	2.02
E. coli	4.73	2.45	2.64	2.00
Pseudomonas sp.	----	3.83	3.50	3.21

The values are all $\log_{10}$ of the mean number per milliliter. Values were not calculated for Serratia because of its erratic occurrence and detectability. By the same token the presence of Pseudomonas in the influent wastewater was known and detected; however, detection by standard plate techniques was hampered due to the difficulty encountered in culturing this genus at standard laboratory incubation temperatures.

The data presented above demonstrate that pond Series I were the most efficient in reducing the total bacterial populations. The slightly increased detention time may have been a contributing factor in this reduction. For total coliform bacteria all three series exhibited similar capabilities; however, Series III exhibited slightly more efficiency in this regard than did I or II. Similarly, Series III performed slightly more efficiently than did Series I or II in reducing E. coli numbers as well as Pseudomonas.

Escherichia coli, Pseudomonas aeruginosa, and Serratia marcescens all exhibited some survival at the three effluent sample stations 5, 7, and 11. Some correlation between the phytoplankton concentrations and the bacterial densities were observed. Pseudomonas appeared in higher nubbers when reduced phytoplankton concentrations were present. E. coli apparently was capable of survival in consistently higher numbers regardless of the phytoplankton concentrations. Serratia was rarely present in consistently large concentrations in the effluents; however, the species inoculated was apparently different from that (or those) present in the wastewater. The periodic increases in Serratia (Tables B-9 and B-10, Appendix B) were observed to be associated with clumps of blue-green algae, a phenomenon which is common to all waste stabilization ponds during the summer and early fall seasons.

The group of organisms which are reported in Tables B-11 and B-12 of Appendix B, the chromogens, includes such genera as Flaveobacterium and Brevibacterium. These organisms were found to be present in wastewater in high concentrations. Very little is known about their physiology, pathogenicity (if any), or their contribution to the overall dieoff of the coliform group. From the data presented in Tables B-11 and B-12 it can be seen that aftergrowth of these bacterial genera did occur in the effluent sampling station zones 5, 7 and 11. The comparatively high numbers of these bacteria may have been responsible, at least in part, for a certain amount of the dieoff of the coliforms and/or test species. This may have occurred by either antibiosis or nutrient competition. At any rate, Flaveobacterium and Brevibacterium will require considerably more research effort before their exact contribution to waste treatment processes can be effectively evaluated.

Field Waste Stabilization Pond Studies

During and preceding the period of investigation with the laboratory waste stabilization ponds, data were obtained on three series of field waste stabilization ponds. These data were taken for a three-month period beginning on June 4 and extending through August 26. A diagram of the field ponds is shown in Figure 4-6. The laboratory waste stabilization ponds described earlier in the text were designed to approximate the detention times and types of systems of the field ponds. Series I was preceded by an anaerobic pretreatment unit which had a volume of 8,900 cubic feet and a detention time of about 4 days. The facultative pond in Series I had a volume of 117,500 cubic feet and a detention time of about 55 days. The small maturation ponds all had volumes of 18,000 cubic feet and detention times of about 8 days. Series II and III facultative ponds had volumes of 126,400 cubic feet and 126,300 cubic feet respectively with detention times of about 51 days and 59 days each. Sample stations were in the vicinities of the numbered areas of the ponds in Figure 4-6. Surface samples were taken one foot below the surface and bottom samples were taken about one foot from the bottom. This was done to avoid excessive concentrations of algae at the surface and excessive amounts of settled sludge at the bottom. Data obtained from these 12 sampling points are presented in detail in Tables B-13 through B-28, Appendix B. Objectives of this phase of the investigation were to compare the efficiencies of the three different types of waste stabilization ponds as to coliform reduction, to compare the efficiencies of the field and laboratory scale ponds, and to attempt massive inocula of selected bacteria for dieoff studies.

Inoculations with laboratory-cultured bacteria occurred on the dates indicated in Table 4-5. The numbers of bacteria which were inoculated into those ponds in certain cases proved to be ineffective as tracer methodology for dieoff studies. Dieoff of the test species was rapid in the field ponds. Calculating by volume of the ponds and daily flow rates from the influent to the sample stations indicated the following dieoff coefficients (as $\log_{10}$ day^{-1}).

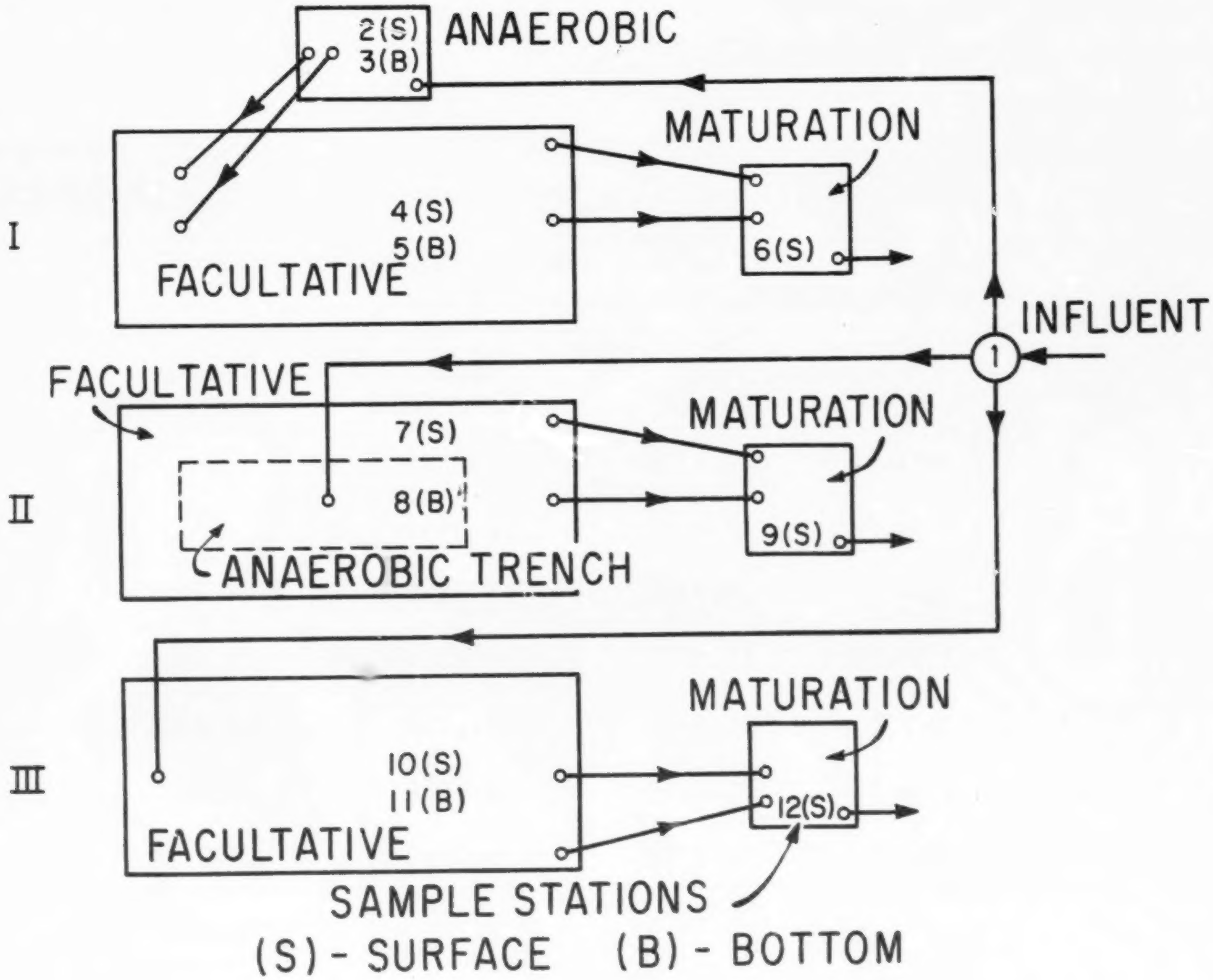
2(S)
3(B)
ANAEROBIC
MATURATION
I
4(S)
5(B)
FACULTATIVE
6(S)
INFLUENT
1
FACULTATIVE
7(S)
MATURATION
II
8(B)
9(S)
ANAEROBIC TRENCH
MATURATION
III
10(S)
11(B)
FACULTATIVE
12(S)
SAMPLE STATIONS
(S) - SURFACE (B) - BOTTOM

FIG. 4-6. SCHEMATIC OF WASTE STABILIZATION PONDS

Table 4-5. Bacteria Inoculated Into Selected Stations of Waste Stabilization Ponds

Station	Date	*Escherichia coli*	*Pseudomonas aeruginosa*	*Serratia marcescens*
2	June 5	13.2265*	14.0592	--
4	June 5	12.9350	12.9351	--
4	June 16	13.7164	14.2812	14.1763
7	June 19	13.9235	14.0214	13.9855
7	July 29	15.0281	15.1146	14.4340
10	June 23	13.4510	14.1565	14.5513
10	July 21	14.0720	14.5761	14.2140

*$\log_{10}$ numbers per ml.

Bacterial Species	Sample Stations		
	#4	#7	#10
Escherichia coli	-1.42	-1.67	-1.21
Pseudomonas aeruginosa	-0.89	-1.10	-0.91
Serratia marcescens	-1.81	-2.02	-.199

These coefficients were, without exception, higher than those found for the laboratory scale ponds, indicating accelerated antibiotic activites. Solar radiation contributed to some extent to those accelerated rates. Data for total bacteria, total coliform bacteria, and phytoplankton populations are shown in Figures 4-7 through 4-9, and Tables 4-6 and 4-7. Those data indicate, as in the case of the laboratory waste stabilization pond studies, that a period of acclimatization was occurring for approximately half the test period of three months. Means of total bacteria, total coliform bacteria and *E. coli* concentrations were calculated for the raw influent (station 1) and the three effluents for the test period. They were as follows (as $\log_{10}$ No./ml).

	Influent	Series I Station 6	Series II Station 9	Series III Station 12
Total Bacteria	7.14	5.04	4.91	5.25
Total Coliform	5.83	1.35	1.31	1.37
E. coli	5.26	0.34	0.78	0.57

These values indicate that reduction of the total bacteria was not as efficient as could be hoped for. Total coliform bacteria were reduced significantly, with the majority of reduction being due to the dieoff of *E. coli*, per se.

Algae which were present in the ponds throughout the test period were predominantly blue-green and green algae, as was the case for the laboratory waste stabilization ponds. Surprisingly high concentrations of algae were found at the surface of the anaerobic pretreatment pond of Series I, contributing, no doubt, to some aerobic activity. There appeared to be a direct relationship between the high phytoplankton concentrations in August and consistently lower coliform counts. Increases in total coliform

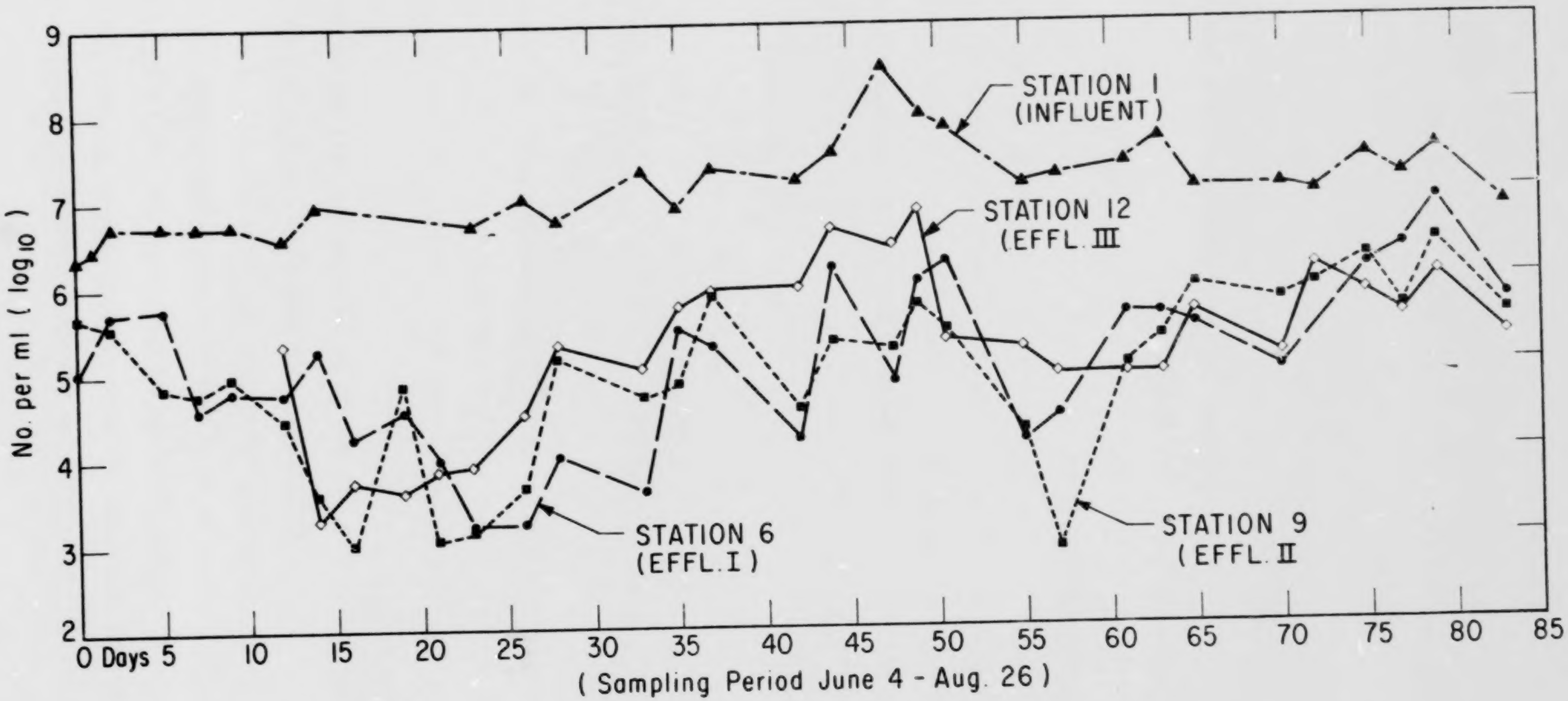

FIG. 4-7. TOTAL BACTERIA IN WASTE STABILIZATION PONDS

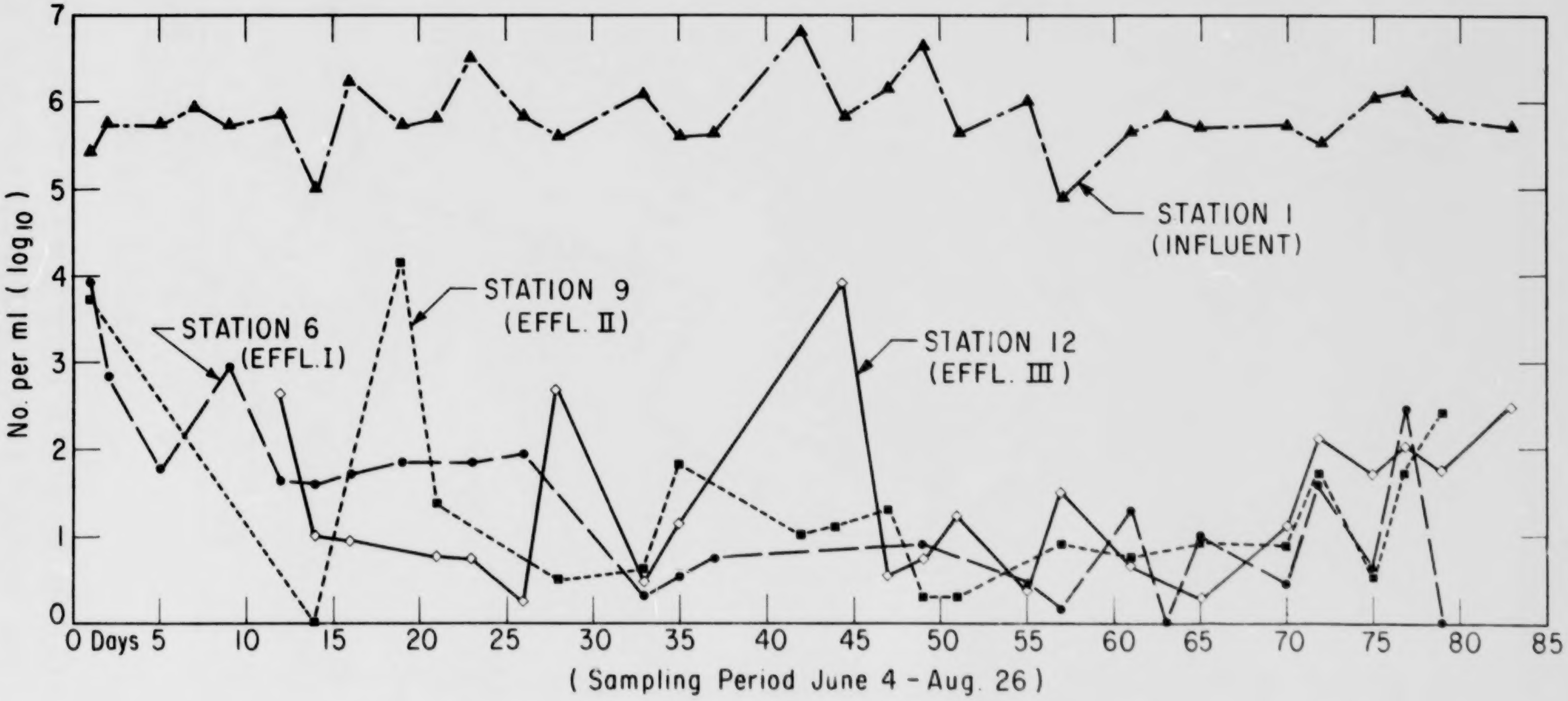

FIG. 4-8. TOTAL COLIFORM IN WASTE STABILIZATION PONDS

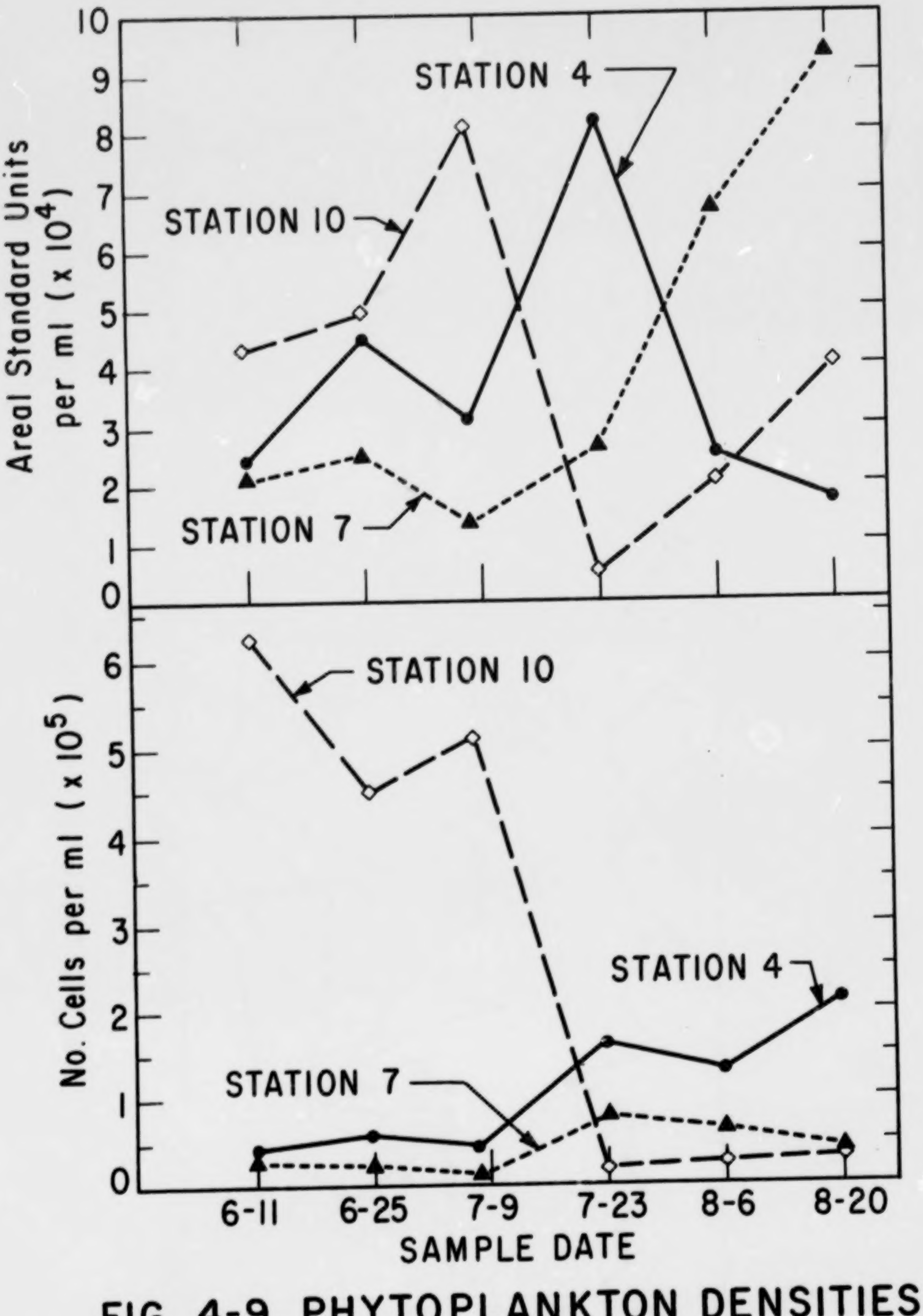

FIG. 4-9. PHYTOPLANKTON DENSITIES IN WASTE STABILIZATION PONDS

Table 4-6. Phytoplankton Found in Waste Stabilization Ponds

Algal Division	Date and Station Number								
	June 11			June 25			July 2		
Station:	4	7	10	4	7	10	4	7	10
Cyanophyta	5,000* (7,500)**	5,500 (4,800)	4,100 (2,500)	4,200 (8,000)	5,000 (5,000)	1,500 (4,700)	7,000 (22,000)	3,400 (3,000)	21,000 (24,500)
Euglenophyta	-0-	-0-	1,500 (1,750)	600 (650)	-0-	-0-	-0-	4,000 (3,000)	6,000) (4,500)
Chrysophyta	-0-	-0-	-0-	-0-	-0-	-0-	-0-	-0-	-0-
Chlorophyta	19,100 (42,500)	15,000 (31,200)	37,500 (612,750)	40,150 (51,350)	20,000 (22,500)	47,500 (445,300)	24,500 (27,000)	6,100 (8,000)	54,000 (488,000)
Total	24,100 (45,000)	20,500 (36,000)	43,100 (617,000)	45,000 (60,000)	25,000 (27,500)	49,000 (450,000)	31,500 (49,000)	13,500 (14,000)	81,000 (517,000)

Table 4-6 Continued

Algal Division	Date and Station Number								
	July 23			August 6			August 20		
Station:	4	7	10	4	7	10	4	7	10
Cyanophyta	72,000 (145,000)	9,000 (17,000)	1,000 (500)	20,000 (8,000)	21,200 (10,000)	15,500 (12,000)	3,000 (2,000)	82,000 (17,000)	-0-
Euglenophyta	-0-	-0-	-0-	1,000 (1,000)	1,500 (1,200)	250 (100)	2,000 (1,000)	-0-	-0-
Chrysophyta	-0-	900 (1,000)	-0-	-0-	-0-	-0-	-0-	-0-	-0-
Chlorophyta	9,000 (20,000)	16,000 (57,000)	3,900 (19,500)	4,000 (123,000)	44,300 (50,800)	5,250 (13,400)	13,000 (207,000)	11,000 (22,000)	40,000 (27,000)
Total	81,000 (165,000)	25,400 (75,000)	4,900((20,000)	25,000 (132,000)	67,000 (62,000)	21,000 (25,500)	17,000 (210,000)	93,000 (39,000)	40,000 (27,000)

*Areal Standard Units of phytoplankton per ml.

**( ) No. of phytoplankton per ml.

Table 4-7. Phytoplankton Concentrations Found In Waste Stabilization Ponds

	Station Number										
Date	2	3	4	5	6	7	8	9	10	11	12
June 11	1,100* (900)**	500 (400)	24,100 (45,000)	22,500 (30,000)	7,000 (13,000)	20,500 (36,000)	17,000 (22,500)	5,500 (11,000)	43,100 (617,000)	11,000 (13,500)	2,700 (3,500)
June 25	2,100 (1,300)	250 (100)	45,000 (60,000)	30,000 (27,000)	6,500 (12,500)	25,000 (27,500)	7,900 (15,000)	4,900 (7,200)	49,000 (450,000)	6,200 (12,500)	3,850 (14,000)
July 2	96,900 (2,001,000)	3,600 (1,009,000)	31,500 (49,000)	28,700 (55,000)	6,400 (17,000)	13,500 (14,000)	4,600 (6,000)	9,800 (23,000)	81,000 (517,000)	31,900 (555,000)	26,000 (380,000)
July 23	16,670 (27,200)	60,000 (80,000)	81,000 (165,000)	88,000 (175,000)	66,000 (285,000)	25,900 (75,000)	2,000 (3,000)	19,700 (21,000)	4,900 (20,000)	5,000 (38,000)	7,850 (43,000)
Aug. 6	13,700 (20,000)	7,500 (13,500)	25,000 (132,000)	22,500 (35,750)	25,500 (38,500)	67,000 (62,000)	11,500 (13,000)	21,500 (30,000)	21,000 (25,500)	13,700 (33,000)	7,000 (11,000)
Aug. 20	17,500 (25,500)	6,500 (11,500)	17,000 (210,000)	19,500 (218,000)	40,000 (67,500)	93,000 (39,000)	21,000 (19,000)	8,100 (13,400)	40,000 (27,000)	1,000 (1,100)	14,000 (17,500)

*Areal Standard Units of phytoplankton per ml.
**( ) No. of phytoplankton per ml.

counts in late August (day 70 on) corresponded closely with the comparatively lower algae counts for the period.

Survival of Pseudomonas sp. in these ponds was apparently very difficult as can be seen by the data in Tables B-22 through B-24. Some carryover and aftergrowth was evident in the maturation pond of Series III (station 12). That particular station had a bloom of Brachionus around July 17 in concentrations of up to 200 per milliliter in the surface waters. Those large numbers should have significantly reduced the bacterial population in that time period; however, no significant reduction was noted. On many occasions the concentration of coliform bacteria were lower in the deeper waters than in the surface waters. It is entirely possible that a greater amount of antibiotic activity was occurring in the deeper waters. Apparently few Serratia sp. were present in the influent wastewater. Those individuals which were present appeared to survive until reaching the facultative or maturation ponds. Some aftergrowth of this genus was observed in the maturation ponds. The chromagens exhibited very low, if any, dieoff in most instances. Their higher numbers may have contributed to the overall comparatively efficient reduction of the coliforms. It is clear that the field waste stabilization ponds were more effective in bacterial reduction than were the laboratory units, so far as the coliform group were concerned.

These investigations on the field pilot waste stabilization ponds and the laboratory ponds were accomplished in conjunction with investigations in progress under grant WTRD 178-01-68, Federal Water Pollution Control Administration, "Design Guides for Selected Wastewater Treatment Processes."

With the information presented by this investigation, remaining efforts in this important area of sanitary engineering should be directed toward establishing: (a) guidelines on amounts of disinfectant necessary to eliminate proportions of enteric bacterial populations to meet effluent specifications; and (b) cost-benefit specifications based on the as yet unknown pathogenicity of other bacterial species found in waste stabilization ponds and related wastewater treatment systems. An undeniable need exists at the present time for this information.

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APPENDIX A

STATISTICS OF ALL LABORATORY AXENIC CULTURE STUDIES

Key to Appendix A tabulated columns:

N = number of data points used in computing that particular regression line

S_H^2 = the variance of data points about the mean of all data points

S_r^2 = the variation of data points about the regression line

S_H^2/S_r^2 = the variance ratio which if referenced against appropriate standard "F" tables would indicate the statistical validity of the data.

b = calculated y-intercept

k = the dieoff (-) or growth (+) rate coefficient of the typical $C_t = C_0\ 10^{kt}$ formulation

R = the multiple correlation coefficient

Table A-1

Reduction statistics of enteric bacteria species with *Anabaena cylindrica*. Series BG-I. Bacteria added to algae when in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	57.966	0.6783	(8.7213± .8873)	(-.0774±.0192)	0.892
Enterobacter aerogenes					
12	227.04	0.2110	(8.3621± .4949)	(-.0854±.0107)	0.970
Escherichia coli					
12	35.917	0.2882	(8.0230± .5784)	(-.0397±.0125)	0.837
Proteus vulgaris					
12	79.048	0.8310	(8.3912± .9822)	(-.1000±.0213)	0.919
Pseudomonas aeruginosa					
13	17.007	0.6349	(8.2665±.9368)	(-.0512±.0241)	0.739
Serratia marcescens					
14	3.1013	1.7802	(6.8402±1.5687)	(-.0366±.0404)	0.341

Table A-2

Reduction statistics of enteric bacteria species with *Anacystis nidulans*. Series BG-I. Bacteria added to algae when in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	78.809	0.4033	(7.9753±.8313)	(-.1145±.0260)	0.940
Enterobacter aerogenes					
12	97.721	0.3406	(7.7923±.7639)	(-.1172±.0239)	0.951
Escherichia coli					
12	116.24	0.3905	(8.1602±.6598)	(-.0796±.0137)	0.936
Proteus vulgaris					
12	111.76	0.3614	(7.6181±.6798)	(-.0899±.0161)	0.941
Pseudomonas aeruginosa					
13	57.109	0.2387	(7.8915±.5883)	(-.0614±.0158)	0.904
Serratia marcescens					
14	83.845	0.1375	(8.3103±.4194)	(-.0480±.0099)	0.923

Table A-3

Reduction statistics of enteric bacteria species with *Gloeocapsa alpicola*. Series BG-I. Bacteria added to algae when in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	36.494	0.9304	(6.5049±1.0184)	(-.0688±.0212)	0.820
Enterobacter aerogenes					
12	79.796	0.1942	(8.1088±.7585)	(-.1356±.0357)	0.964
Escherichia coli					
12	44.764	0.5826	(7.7978±.9192)	(-.0849±.0246)	0.882
Proteus vulgaris					
12	8.3744	1.4614	(8.3445±2.0807)	(-.1205±.0980)	0.736
Pseudomonas aeruginosa					
13	52.261	0.1064	(7.7185±.5751)	(-.0484±.0157)	0.946
Serratia marcescens					
14	20.3386	0.1045	(8.0011±.7897)	(-.0466±.0301)	0.910

Table A-4

Reduction statistics of enteric bacteria species with *Oscillatoria chalybia*. Series BGI. Bacteria added to algae when in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	22.511	2.0704	(6.5580±1.6270)	(-.0966±.0386)	0.763
Enterobacter aerogenes					
12	6.1434	1.5995	(6.1868±1.6554)	(-.0637±.0518)	0.551
Escherichia coli					
12	31.303	0.6905	(6.4209±1.2376)	(-.1255±.0478)	0.887
Proteus vulgaris					
12	136.60	0.2356	(6.9627±.6353)	(-.1153±.0199)	0.965
Pseudomonas aeruginosa					
13	64.500	0.4957	(8.4764±.9215)	(-.1149±.0288)	0.928
Serratia marcescens					
14	109.20	0.2419	(7.4993±.5193)	(-.0607±.0108)	0.932

Table A-5

Reduction statistics of enteric bacteria species with Oscillatoria formosa. Series BGI. Bacteria added to algae when in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	216.93	0.1511	(8.6823±.5790)	(-.1546±.0224)	0.982
Enterobacter aerogenes					
12	14.656	3.7099	(7.3945±2.0752)	(-.0910±.0450)	0.677
Escherichia coli					
12	17.347	2.8252	(7.1810±1.8109)	(-.0864±.0393)	0.712
Proteus vulgaris					
12	12.962	4.7076	(6.5127±2.3376)	(-.0964±.0507)	0.649
Pseudomonas aeruginosa					
13	15.716	2.6409	(7.1070±1.7509)	(-.0795±.0379)	0.692
Serratia marcescens					
14	65.150	0.4653	(7.6482±.7349)	(-.0679±.0159)	0.903

Table A-6

Reduction statistics of enteric bacteria species with Phormidium faveolarum. Series BGI. Bacteria added to algae when in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	14.979	3.3952	(6.9144±1.9852)	(-.0880±.0431)	0.681
Enterobacter aerogenes					
12	6.9355	3.8784	(5.0351±2.1218)	(-.0640±.0460)	0.498
Escherichia coli					
12	9.8227	3.9133	(5.0539±2.3258)	(-.0966±.0599)	0.621
Proteus vulgaris					
12	25.151	1.7319	(6.2764±1.9600)	(-.1782±.0757)	0.863
Pseudomonas aeruginosa					
13	71.313	0.3654	(7.1779±.9003)	(-.1378±.0348)	0.947
Serratia marcescens					
14	4.9669	1.6954	(6.8908±2.2411)	(-.0999±.1055)	0.623

Table A-7

Reduction statistics of bacterial contaminants of *Anabaena cylindrica* during series BG-I with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	0.7611	0.3184	(9.3911±1.3784)	(-.0157±.0526)	0.276
Enterobacter aerogenes					
12	12.004	0.0249	(9.7528±.3856)	(-.0175±.0147)	0.857
Escherichia coli					
12	0.6891	0.1714	(9.6374±2.3863)	(-.0173±.1320)	0.408
Proteus vulgaris					
11	0.8337	0.0605	(9.7131±.4338)	(-.0046±.0118)	0.217
Pseudomonas aeruginosa					
12	45.860	0.0051	(9.3210±.1736)	(-.0154±.0066)	0.958
Serratia marcescens					
12	0.7622	0.0224	(9.6187±.2641)	(-.0027±.0072)	0.203

Table A-8

Reduction statistics of bacterial contaminants of *Anacystis nidulans* during series BG-I with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	0.1918	0.3267	(8.4975±1.0079)	(.0051±.0275)	0.060
Enterobacter aerogenes					
12	5.2952	0.0113	(9.7894±.2597)	(-.0078±.0099)	0.726
Escherichia coli					
12	6.5719	0.0472	(9.4214±.3833)	(-.0114±.0105)	0.686
Proteus vulgaris					
12	0.8462	0.8866	(8.9210±1.6604)	(-.0177±.0453)	0.220
Pseudomonas aeruginosa					
12	2.0035	1.6987	(9.9329±3.1842)	(-.0589±.1216)	0.500
Serratia marcescens					
12	34.179	0.0150	(9.2648±.2163)	(-.0147±.0059)	0.919

Table A-9

Reduction statistics of bacterial contaminants of *Gloeocapsa alpicola* during series BG-I with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	12.7660	0.0290	(7.6995±.4162)	(-.0194±.0159)	0.864
Enterobacter aerogenes					
12	355.73	0.0246	(8.9674±.2769)	(-.0606±.0075)	0.992
Escherichia coli					
12	20.605	0.3061	(8.2531±1.3517)	(-.0802±.0516)	0.911
Proteus vulgaris					
12	25.218	0.2025	(8.5118±.7937)	(-.0462±.0217)	0.894
Pseudomonas aeruginosa					
12	52.609	0.1064	(7.7185±.5751)	(-.0484±.0157)	0.946
Serratia marcescens					
12	20.338	0.1050	(8.0011±.7897)	(-.0466±.0301)	0.910

Table A-10

Reduction statistics of bacterial contaminants of *Oscillatoria chalybia* during series BG-I with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	43.578	0.1113	(5.1798±.8152)	(-.0704±.0311)	0.956
Enterobacter aerogenes					
12	44.1414	0.0632	(5.9133±.4432)	(-.0341±.0121)	0.936
Escherichia coli					
13	0.5146	2.9367	(5.6901±4.1866)	(-.0393±.1598)	0.205
Proteus vulgaris					
11	9.7113	0.5394	(6.9309±1.2951)	(-.0468±.0353)	0.764
Pseudomonas aeruginosa					
12	6.3348	0.0851	(7.8599±.7127)	(-.0234±.0272)	0.760
Serratia marcescens					
12	13.609	0.0587	(8.8888±.5918)	(-.0285±.0226)	0.872

Table A-11

Reduction statistics of bacterial contaminants of *Oscillatoria formosa* during series BG-I with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	48.049	0.1016	(9.6125±.5447)	(-.0410±.0139)	0.941
Enterobacter aerogenes					
12	11.688	0.1919	(9.7453±.7484)	(-.0278±.0191)	0.796
Escherichia coli					
9	4.8621	0.1967	(9.8661±.7578)	(-.0182±.0194)	0.618
Proteus vulgaris					
12	7.222	0.0850	(9.6902±.7123)	(-.0250±.0272)	0.783
Pseudomonas aeruginosa					
11	21.177	0.2511	(10.0314±.8560)	(-.0428±.0219)	0.876
Serratia marcescens					
11	18.190	0.1096	(9.7702±.8088)	(-.0451±.0309)	0.901

Table A-12

Reduction statistics of bacterial contaminants of *Phormidium faveolarum* during series BG-I with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	0.2333	0.1059	(7.2847±.5559)	(.0029±.0142)	0.072
Enterobacter aerogenes					
12	16.099	0.2447	(9.0394±.8451)	(-.0368±.0216)	0.843
Escherichia coli					
12	11.352	0.0985	(8.7132±.5361)	(-.0196±.0137)	0.791
Proteus vulgaris					
12	3.2495	0.6308	(9.0526±1.3569)	(-.0266±.0347)	0.520
Pseudomonas aeruginosa					
12	27.377	0.0374	(8.7549±.3303)	(-.0188±.0084)	0.901
Serratia marcescens					
12	23.895	0.1088	(8.7565±.8059)	(-.0515±.0308)	0.923

Table A-13

Reduction statistics of enteric bacteria species with *Anabaena cylindrica*.
Series BG-II. Bacteria and algae inoculated within
twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	6.459	4.5436	(5.4092±2.0979)	(-.0582±.0419)	0.418
Enterobacter aerogenes					
9	7.704	4.2371	(5.6137±2.2365)	(-.0816±.0557)	0.524
Escherichia coli					
9	16.612	3.8936	(6.9423±2.1439)	(-.1132±.0534)	0.697
Proteus vulgaris					
12	16.439	3.1431	(5.6887±1.6783)	(-.0687±.0307)	0.622
Pseudomonas aeruginosa					
12	14.83	0.3628	(7.3219±.5702)	(-.0702±.0104)	0.937
Serratia marcescens					
12	18.062	2.0567	(5.8236±1.3576)	(-.0583±.0249)	0.644

Table A-14

Reduction statistics of enteric bacteria species with *Anacystis nidulans*.
Series BG-II. Bacteria and algae inoculated within
twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	15.144	0.9024	(6.0191±1.1080)	(-.0640±.0320)	0.716
Enterobacter aerogenes					
12	13.014	1.6865	(5.3492±1.2294)	(-.0448±.0225)	0.565
Escherichia coli					
12	52.405	0.7412	(5.6975±.8150)	(-.0596±.0149)	0.840
Proteus vulgaris					
10	4.2597	5.0709	(4.4056±2.2207)	(-.0522±.0470)	0.347
Pseudomonas aeruginosa					
11	40.969	0.5410	(7.0880±.7078)	(-.0474±.0136)	0.820
Serratia marcescens					
12	6.6982	3.1234	(5.7883±1.6731)	(-.0437±.0306)	0.401

Table A-15

Reduction statistics of enteric bacteria species with *Gloeocapsa alpicola*. Series BG-II. Bacteria and algae inoculated within twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	23.541	2.7722	(6.0652±1.6387)	(-.0868±.0328)	0.723
Enterobacter aerogenes					
12	30.543	1.9952	(5.5931±1.3902)	(-.0838±.0278)	0.772
Escherichia coli					
12	27.325	0.7285	(5.0102±.8524)	(-.0494±.0176)	0.773
Proteus vulgaris					
12	11.356	3.6999	(5.1014±1.9798)	(-.0794±.0438)	0.587
Pseudomonas aeruginosa					
12	27.164	2.4549	(6.4011±1.4833)	(-.0781±.0272)	0.731
Serratia marcescens					
12	36.312	1.0680	(5.4419±.9783)	(-.0596±.0179)	0.784

Table A-16

Reduction statistics of enteric bacteria species with *Oscillatoria chalybia*. Series BG-II. Bacteria and algae inoculated within twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	18.249	2.7641	(5.4482±1.6169)	(-.0761±.0327)	0.670
Enterobacter aerogenes					
12	46.006	0.6161	(5.4358±.7633)	(-.0571±.0154)	0.836
Escherichia coli					
12	44.993	0.751	(8.2274±1.3374)	(-.1986±.0631)	0.918
Proteus vulgaris					
12	21.521	1.7036	(5.5990±1.5143)	(-.1074±.0450)	0.782
Pseudomonas aeruginosa					
12	41.208	0.4405	(6.4460±.7140)	(-.0614±.0181)	0.855
Serratia marcescens					
12	38.386	1.4187	(6.7981±1.2119)	(-.0905±.0272)	0.827

Table A-17

Reduction statistics of enteric bacteria species with *Oscillatoria formosa*. Series BG-II. Bacteria and algae inoculated within twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	15.581	1.3746	(6.8638±2.8644)	(-.2957±.2187)	0.886
Enterobacter aerogenes					
12	7.3510	0.6556	(6.8156±4.6670)	(-.2218±.5164)	0.880
Escherichia coli					
12	69.975	0.3625	(8.0650±1.0976)	(-.2275±.0640)	0.959
Proteus vulgaris					
12	27.448	0.8883	(7.9273±1.781)	(-.2231±.1002)	0.901
Pseudomonas aeruginosa					
12	27.483	2.2444	(6.7767±1.4182)	(-.0751±.0260)	0.733
Serratia marcescens					
12	60.798	0.9046	(6.5623±.9004)	(-.0709±.0165)	0.859

Table A-18

Reduction statistics of enteric bacteria species with *Phormidium faveolarum*. Series BG-II. Bacteria and algae inoculated within twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	35.483	1.4550	(5.5216±1.2415)	(-.0880±.0275)	0.816
Enterobacter aerogenes					
12	9.3918	1.8409	(4.7479±1.4742)	(-.0594±.0367)	0.573
Escherichia coli					
12	10.2230	3.2589	(5.1780±1.7090)	(-.0552±.0313)	0.505
Proteus vulgaris					
12	9.5460	1.2614	(5.7131±2.0473)	(-.1567±.1194)	0.761
Pseudomonas aeruginosa					
12	54.309	0.5361	(6.9504±1.3347)	(-.2437±.0778)	0.948
Serratia marcescens					
12	10.628	2.1393	(4.4701±1.3846)	(-.0456±.0254)	0.515

Table A-19

Reduction statistics of bacterial contaminants of Anabaena cylindrica during series BG-II with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
13	44.545	0.0424	(8.9466±.2816)	(-.0187±.0054)	0.881
Enterobacter aerogenes					
12	5.8129	2.3427	(8.2424±.6616)	(-.0159±.0128)	0.492
Escherichia coli					
12	2.0875	0.3648	(8.6235±1.0190)	(-.0175±.0258)	0.343
Proteus vulgaris					
12	0.7149	0.6621	(8.1277±1.1123)	(-.0094±.0215)	0.106
Pseudomonas aeruginosa					
13	5.9898	0.2366	(8.6531±.7359)	(-.0198±.0163)	0.545
Serratia marcescens					
13	10.141	0.1349	(8.8000±.6195)	(-.0234±.0157)	0.717

Table A-20

Reduction statistics of bacterial contaminants of Anacystis nidulans during series BG-II with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
13	32.316	0.0324	(8.5258±.2666)	(-.0197±.0074)	0.890
Enterobacter aerogenes					
12	14.086	0.1689	(8.8843±.5258)	(-.0213±.0114)	0.738
Escherichia coli					
12	3.7262	0.3657	(8.3519±.7735)	(-.0161±.0168)	0.427
Proteus vulgaris					
12	16.150	0.2533	(8.5266±.6437)	(-.0279±.0140)	0.763
Pseudomonas aeruginosa					
12	0.3013	0.4104	(7.6799±.9494)	(-.0068±.0263)	0.070
Serratia marcescens					
12	26.612	0.1054	(8.4857±.4812)	(-.0323±.0133)	0.869

Table A-21

Reduction statistics of bacterial contaminants of *Gloeocapsa alpicola* during Series BG-II with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
13	3.2359	0.2076	(7.9638±.6747)	(-.0110±.0130)	0.447
Enterobacter aerogenes					
12	13.108	0.1672	(8.1383±.6055)	(-.0198±.0117)	0.766
Escherichia coli					
12	5.7870	0.3437	(8.8736±1.0537)	(-.0271±.0265)	0.658
Proteus vulgaris					
12	8.1439	0.1337	(8.5076±.5415)	(-.0139±.0104)	0.671
Pseudomonas aeruginosa					
12	6.4973	0.0826	(7.9595±.5167)	(-.0141±.0130)	0.684
Serratia marcescens					
12	16.613	0.1527	(8.2359±.5786)	(-.0213±.0111)	0.806

Table A-22

Reduction statistics of bacterial contaminants of *Oscillatoria chalybia* during series BG-II with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	24.990	0.0192	(7.6573±.2627)	(-.0110±.0052)	0.893
Enterobacter aerogenes					
12	24.106	0.0310	(7.7886±.3343)	(-.0138±.0066)	0.889
Escherichia coli					
12	12.224	0.0996	(8.3854±.5177)	(-.0143±.0087)	0.753
Proteus vulgaris					
12	0.2826	0.1938	(7.4938±.8353)	(-.0037±.0165)	0.086
Pseudomonas aeruginosa					
12	0.0174	0.0899	(7.1715±.5690)	(.0006 ±.0112)	0.006
Serratia marcescens					
12	0.7189	0.1208	(7.3817±.6596)	(-.0047±.0130)	0.193

Table A-23

Reduction statistics of bacterial contaminants of *Oscillatoria formosa* during series BG-II with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	0.0758	0.2549	(7.7277±.7207)	(-.0018±.0139)	0.019
Enterobacter aerogenes					
12	0.4654	0.3258	(7.7450±.8148)	(-.0051±.0158)	0.104
Escherichia coli					
12	3.7691	0.2270	(8.0401±.6801)	(-.0120±.0132)	0.485
Proteus vulgaris					
12	36.3346	0.0251	(8.0503±.2259)	(-.0124±.0044)	0.901
Pseudomonas aeruginosa					
12	1.5084	0.1332	(7.7167±.5209)	(-.0058±.0101)	0.274
Serratia marcescens					
12	1.1490	1.4257	(7.8396±.9224)	(-.0129±.0352)	0.365

Table A-24

Reduction statistics of bacterial contaminants of *Phormidium faveolarum* during series BG-II with enteric bacteria.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	3.8100	0.1202	(7.7007±2.0520)	(-.0195±.0630)	0.792
Enterobacter aerogenes					
12	7.2137	0.1341	(7.8364±.9043)	(-.0174±.0189)	0.783
Escherichia coli					
12	10.6154	0.2067	(8.1078±1.1228)	(-.0262±.0235)	0.841
Proteus vulgaris					
12	2.8200	0.2284	(7.7159±1.1805)	(-.0142±.0247)	0.585
Pseudomonas aeruginosa					
12	2.6126	0.0269	(6.2282±.4053)	(.0047±.0085)	0.566
Serratia marcescens					
12	2.8029	1.2669	(7.4335±6.6603)	(-.0542±.2044)	0.737

Table A-25

Growth statistics of bacteria found in axenic algae cultures. Series BG-III. Controls.

N	S_H^2/S_r^2	S_r^2	b	k	R
Anabaena cylindrica					
11	3.6350	5.0979	(3.7588±3.2707)	(.0557±.0622)	0.476
Anacystis nidulans					
10	2.4846	5.8969	(3.5555±3.5177)	(.0495±.0669)	0.383
Gloeocapsa alpicola					
12	14.403	1.4564	(1.8642±1.7482)	(.0592±.0333)	0.783
Oscillatoria chalybia					
12	6.0545	2.3982	(3.9318±2.2433)	(.0493±.0427)	0.602
Oscillatoria formosa					
12	3.0423	3.4652	(4.4801±2.6966)	(.0420±.0513)	0.432
Phormidium faveolarum					
12	4.5869	2.7219	(4.1262±2.3899)	(.0457±.0455)	0.534

Table A-26

Growth statistics of axenic culture of Anabaena cylindrica with enteric bacteria during series BG-I run. Series BG-IV. Results based on mg/l dry weight.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	6.2784	0.0564	(1.4545±.5655)	(.0117±.0099)	0.611
Enterobacter aerogenes					
11	8.6185	0.0342	(1.7229±.4403)	(.0107±.0079)	0.683
Escherichia coli					
11	0.4352	0.1098	(1.8928±.7893)	(.0043±.0139)	0.098
Proteus vulgaris					
11	2.9086	0.1148	(1.4919±.8073)	(.0114±.0143)	0.421
Pseudomonas aeruginosa					
11	10.575	0.0580	(1.2489±.5738)	(.0155±.0101)	0.725
Serratia marcescens					
11	12.066	0.0493	(1.2862±.5288)	(.0152±.0093)	0.751

Table A-27

Growth statistics of axenic culture of *Anacystis nidulans* with enteric bacteria during series BG-I run. Series BG-IV. Results based on mg/l dry weight.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	2.7980	0.1230	(1.4449±.8355)	(.0116±.0148)	0.411
Enterobacter aerogenes					
11	5.5746	0.0575	(1.1917±.5714)	(.0112±.0101)	0.582
Escherichia coli					
11	67.451	0.0065	(1.602±.1919)	(.0131±.0034)	0.944
Proteus vulgaris					
11	2.7309	0.0389	(1.5815±.4699)	(.0064±.0083)	0.406
Pseudomonas aeruginosa					
11	1.5814	0.1693	(1.3273±.9802)	(.0102±.0173)	0.283
Serratia marcescens					
11	6.3484	0.0513	(1.3299±.5398)	(.0113±.0095)	0.613

Table A-28

Growth statistics of axenic culture of *Gloeocapsa alpicola* with enteric bacteria during series BG-I run. Series BG-IV. Results based on mg/l dry weight.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	1.9229	0.0928	(1.6702±.7257)	(.0083±.0128)	0.324
Enterobacter aerogenes					
11	4.2808	0.0297	(1.9153±.4106)	(.0070±.0072)	0.517
Escherichia coli					
11	5.7088	0.0326	(1.7957±.4299)	(.0085±.0076)	0.588
Proteus vulgaris					
11	0.6004	0.0673	(2.1234±.6180)	(.0039±.0109)	0.130
Pseudomonas aeruginosa					
11	4.3449	0.0140	(2.0407±.2821)	(.0049±.0050)	0.521
Serratia marcescens					
11	17.924	0.0203	(1.5473±.3396)	(.0119±.0060)	0.817

Table A-29

Growth statistics of axenic culture of Oscillatoria chalybia with enteric bacteria during series BG-I run. Series BG-IV. Results based on mg/l dry weight.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	8.9103	0.0203	(1.5632±.3391)	(.0084±.0059)	0.690
Enterobacter aerogenes					
11	8.5231	0.0134	(1.6601±.2759)	(.0067±.0048)	0.681
Escherichia coli					
11	0.7271	0.0860	(1.9015±.6988)	(.0049±.0123)	0.154
Proteus vulgaris					
11	26.7943	0.0343	(1.7519±.4414)	(.0189±.0078)	0.870
Pseudomonas aeruginosa					
11	10.575	0.0580	(1.2489±.5738)	(.0155±.0101)	0.725
Serratia marcescens					
11	12.066	0.0493	(1.2862±.5288)	(.0152±.0093)	0.751

Table A-30

Growth statistics of axenic culture of Oscillatoria formosa with enteric bacteria during series BG-I run. Series BG-IV. Results based on mg/l dry weight.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	12.215	0.1702	(.6819±.9829)	(.0285±.0174)	0.753
Enterobacter aerogenes					
11	22.734	0.0742	(1.0015±.6489)	(.0256±.0115)	0.850
Escherichia coli					
13	9.3189	0.1202	(1.1998±.8261)	(.0209±.0146)	0.699
Proteus vulgaris					
12	2.9976	0.2196	(1.3486±1.1164)	(.0160±.0197)	0.428
Pseudomonas aeruginosa					
11	1.4971	0.1744	(1.5118±.9948)	(.0101±.0176)	0.272
Serratia marcescens					
11	3.3157	0.1967	(1.3871±1.0566)	(.0159±.0187)	0.453

Table A-31

Growth statistics of axenic culture of *Phormidium faveolarum* with enteric bacteria during series BG-I run. Series BG-IV. Results based on mg/l dry weight.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	0.0620	0.0763	(2.1178±.6583)	(.0013±.0116)	0.015
Enterobacter aerogenes					
12	0.0144	0.0504	(2.0452±.5349)	(.0005±.0094)	0.004
Escherichia coli					
12	0.4124	0.0444	(2.1646±.5023)	(-.0027±.0088)	0.093
Proteus vulgaris					
12	0.4748	0.1242	(1.6443±.8396)	(.0048±.0148)	0.106
Pseudomonas aeruginosa					
12	0.1167	0.0785	(2.0089±.6676)	(-.0019±.0118)	0.028
Serratia marcescens					
12	1.1185	0.0804	(2.3023±.6755)	(-.0059±.0119)	0.218

Table A-32

Reduction statistics of enteric bacteria species with algal contaminant *Brevibacterium*, Series BG-V.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	11.847	1.3542	(6.7687±1.5672)	(-.0513±.0300)	0.703
Enterobacter aerogenes					
12	61.275	2.4412	(8.3011±.6642)	(-.0494±.0127)	0.924
Escherichia coli					
12	15.235	0.2289	(8.4366±.6432)	(-.0755±.0123)	0.968
Proteus vulgaris					
12	115.08	0.4806	(8.4680±.9319)	(-.0951±.0178)	0.958
Pseudomonas aeruginosa					
12	28.100	1.2490	(7.6973±1.7243)	(-.1011±.0407)	0.875
Serratia marcescens					
12	55.697	0.427	(8.6455±.8787)	(-.0624±.0168)	0.918

Table A-33

Reduction statistics of enteric bacteria species with algal contaminant, Flaveobacterium, Series BG-V.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	19.699	1.0269	(8.1993±2.4757)	(-.1437±.0945)	0.908
Enterobacter aerogenes					
12	188.33	0.0543	(8.0086±1.3435)	(-.1616±.0743)	0.995
Escherichia coli					
12	21.881	1.4051	(8.1903±1.8289)	(-.0947±.0431)	0.845
Proteus vulgaris					
12	136.43	0.3321	(10.0665±1.0505)	(-.1520±.0306)	0.978
Pseudomonas aeruginosa					
12	22.289	0.9558	(7.6614±1.7822)	(-.1042±.0519)	0.881
Serratia marcescens					
12	16.282	1.6677	(6.5629±1.7360)	(-.0666±.0333)	0.765

Table A-34

Reduction statistics of single species of enteric bacteria in presence of mixed cultures of six species of blue-green algae. Series BG-VIII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
13	15.939	1.4918	(7.7778±7.0397)	(-.2463±.3895)	0.941
Enterobacter aerogenes					
12	6.2346	3.5258	(8.1472± 10.8224)	(-.2368±.5988)	0.862
Escherichia coli					
13	0.3556	2.9060	(7.0455±7.1646)	(-.1666±.1590)	0.824
Proteus vulgaris					
13	5.4117	4.1432	(8.1208±11.7319)	(-.2392±.6491)	0.844
Pseudomonas aeruginosa					
12	10.783	2.7206	(7.0364±4.0296)	(-.1730±.1538)	0.843
Serratia marcescens					
12	4.2473	5.0527	(6.4926±5.4915)	(-.1479±.2096)	0.679

Table A-35

Reduction statistics of mixed enteric bacteria in presence of mixed cultures of six species of blue-green algae. Series BG-IX.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	14.056	1.8856	(6.7616±7.9146)	(-.2600±.4379)	0.933
Enterobacter aerogenes					
11	5.9160	4.9779	(6.8051±12.8595)	(-.2741±.7115)	0.855
Escherichia coli					
11	0.0786	19.409	(4.1152±19.6689)	(-.1081±2.4333)	0.073
Proteus vulgaris					
11	4.3410	5.6157	(6.2648±12.6453)	(-.1632±.4947)	0.813
Pseudomonas aeruginosa					
11	13.145	1.7591	(6.9496±3.2402)	(-.1536±.1237)	0.868
Serratia marcescens					
11	36.931	0.5183	(7.0223±1.7588)	(-.1397±.0671)	0.948
Total* of all 6 enterics					
11	20.528	1.1271	(7.5912±2.5936)	(-.1536±.0990)	0.911

* Data for the total number of enteric bacteria, not sum of individual statistical results for each species.

Table A-36

Reduction statistics of enteric bacteria species with *Ankistrodesmus braunii*. Series G-I. Bacteria added to algae when in mid-log phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
13	59.850	0.45717	(7.0994±.9669)	(-.0701±.0193)	0.937
Enterobacter aerogenes					
14	89.272	0.36307	(8.2215±.8617)	(-.0764±.0172)	0.957
Escherichia coli					
14	7.1998	1.0295	(6.7241±1.4509)	(-.0365±.0290)	0.643
Proteus vulgaris					
11	38.053	0.35906	(6.9891±1.0566)	(-.0756±.0288)	0.927
Pseudomonas aeruginosa					
12	42.717	0.29243	(7.9607±1.3211)	(-.1129±.0504)	0.955
Serratia marcescens					
12	11.093	0.82891	(9.1086±1.6055)	(-.0620±.0438)	0.787

Table A-37

Reduction statistics of enteric bacteria species with *Chlorella pyrenoidosa*. Series G-I. Bacteria added to algae when in mid-log phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
13	447.65	.05507	(8.1764±.5733)	(-.1586±.0219)	0.995
Enterobacter aerogenes					
14	9.3163	2.6364	(7.7223±2.8633)	(-.1013±.0781)	0.756
Escherichia coli					
14	19.113	1.1338	(6.6677±1.5227)	(-.0624±.0305)	0.827
Proteus vulgaris					
11	23.078	1.3761	(8.2918±2.8659)	(-.1800±.1094)	0.920
Pseudomonas aeruginosa					
12	9.1331	2.5412	(8.0134±2.8111)	(-.0985±.0767)	0.753
Serratia marcescens					
12	17.501	0.9995	(7.0495±1.7629)	(-.0855±.0481)	0.854

Table A-38

Reduction statistics of enteric bacteria species with *Chlorella vulgaris*. Series G-I. Bacteria added to algae when in mid-log phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	8.5477	1.8059	(7.1179±3.2831)	(-.1255±.1253)	0.810
Enterobacter aerogenes					
12	6.7731	3.1823	(7.1567±3.1458)	(-.0949±0.0858)	0.693
Escherichia coli					
12	4.2474	2.8177	(6.5618±2.4005)	(-.0464±0.0480)	0.515
Proteus vulgaris					
12	6.2347	2.0976	(8.1411±8.3476)	(-.1826±.4619)	0.862
Pseudomonas aeruginosa					
12	19.549	1.4598	(8.1344±2.1307)	(-.1092±.0581)	0.867
Serratia marcescens					
12	12.080	1.8887	(7.9809±3.0490)	(-.0651±.0547)	0.858

Table A-39

Reduction statistics of enteric bacteria species with *Scenedesmus obliquus*. Series G-I. Bacteria added to algae when in mid-log phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	7.1587	1.6250	(6.1286±1.8230)	(-.0458±.0365)	0.642
Enterobacter aerogenes					
12	26.533	0.6903	(6.9105±1.1881)	(-.0574±.0238)	0.869
Escherichia coli					
12	40.527	0.2004	(7.4914±0.7894)	(-.0583±.0215)	0.931
Proteus vulgaris					
12	17.165	1.1059	(6.9057±1.8545)	(-.0891±.0506)	0.851
Pseudomonas aeruginosa					
12	26.296	0.5495	(7.9135±1.3072)	(-.0777±.0357)	0.898
Serratia marcescens					
12	41.964	0.5409	(8.8431±1.2969)	(-.0974±.0354)	0.933

Table A-40

Reduction statistics of enteric bacteria species with *Ankistrodesmus braunii*. Series G-II. Bacteria and algae inoculated within twenty-four hours of one another

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	154.98	0.19994	(8.7434±2.4481)	(-.0857±.0435)	0.994
Enterobacter aerogenes					
12	17.603	1.5360	(8.5471±6.7856)	(-.0801±.1205)	0.946
Escherichia coli					
12	4.5853	2.7007	(7.8609±8.9976)	(-.0542±.1598)	0.821
Proteus vulgaris					
12	1.8897	12.398	(7.2077±19.278)	(-.0745±.3424)	0.654
Pseudomonas aeruginosa					
12	1765.9	0.01860	(8.9704±.8609)	(-.1977±.0297)	0.999
Serratia marcescens					
12	1891.1	0.01366	(9.7349±.7378)	(-.1753±.0254)	0.999

Table A-41

Reduction statistics of enteric bacteria species with *Chlorella pyrenoidosa*. Series G-II. Bacteria and algae inoculated within twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	24.159	0.9386	(7.9536±5.3042)	(-.0743±.0942)	0.960
Enterobacter aerogenes					
12	2899.2	0.0046	(9.2054±.4299)	(-.1265±.0148)	0.999
Escherichia coli					
12	12.115	2.0249	(7.7306±7.7908)	(-.0763±.1384)	0.924
Proteus vulgaris					
12	69.343	0.1855	(7.0642±2.3582)	(-.0552±.0419)	0.986
Pseudomonas aeruginosa					
12	10.876	1.2791	(8.0599±6.1921)	(-.0574±.1100)	0.916
Serratia marcescens					
12	19.603	0.6550	(8.2736±4.4309)	(-.0552±.0787)	0.951

Table A-42

Reduction statistics of enteric bacteria species with *Chlorella vulgaris*. Series G-II. Bacteria and algae inoculated within twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	2125.2	0.00398	(5.9987±.3983)	(-.1003±.0137)	0.999
Enterobacter aerogenes					
12	2156.9	0.00505	(7.5548±.4487)	(-.1138±.0155)	0.999
Escherichia coli					
12	4.0005	1.7486	(5.6424±7.2399)	(-.0407±.1286)	0.800
Proteus vulgaris					
12	2362.9	0.0065	(8.1123±.5091)	(-.1352±.0176)	0.999
Pseudomonas aeruginosa					
12	4933.7	0.0062	(7.4898±.4954)	(-.1901±.0171)	0.999
Serratia marcescens					
12	1781.4	0.0046	(6.6976±.4292)	(-.0989±.0148)	0.999

Table A-43

Reduction statistics of enteric bacteria species with *Scenedesmus obliquus*. Series G-II. Bacteria and algae inoculated within twenty-four hours of one another.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	847.23	0.02134	(7.4529±.7997)	(-.0655±.0142)	0.998
Enterobacter aerogenes					
12	5911.5	0.00448	(8.3051±.3663)	(-.0792±.0065)	0.999
Escherichia coli					
12	29.286	0.42066	(6.7184±3.5509)	(-.0541±.0631)	0.967
Proteus vulgaris					
12	1821.5	0.00183	(6.4763±.2699)	(-.0629±.0093)	0.999
Pseudomonas aeruginosa					
12	2.1169	4.3286	(6.6961±11.3908)	(-.0466±.2023)	0.679
Serratia marcescens					
12	7.6209	1.3397	(7.5169±6.3371)	(-.0492±.1126)	0.884

Table A-44

Reduction statistics of single species of enteric bacteria in presence of mixed cultures of four species of green algae. Series G-VIII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	47.994	0.5460	(7.5188±1.8052)	(-.1635±.0689)	0.959
Enterobacter aerogenes					
12	145.79	0.1437	(6.6479±.9259)	(-.1462±.0353)	0.986
Escherichia coli					
12	23.382	0.6871	(6.8582±2.0250)	(-.1280±.0773)	0.921
Proteus vulgaris					
12	7.9498	1.8035	(7.3760±7.7403)	(-.1912±.4282)	0.888
Pseudomonas aeruginosa					
12	71.674	0.4158	(8.1735±1.5754)	(-.1744±.0601)	0.973
Serratia marcescens					
12	59.165	0.3693	(7.2299±1.4847)	(-.1493±.0567)	0.967

Table A-45

Reduction statistics of mixed enteric bacteria in presence of mixed cultures of four species of green algae. Series G-IX.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	4.6588	1.1638	(6.6735±6.2179)	(-.1176±.3440)	0.823
Enterobacter aerogenes					
13	2.2508	0.7552	(6.9674±5.0089)	(-.2082±.2771)	0.957
Escherichia coli					
13	17.089	1.1520	(7.3603±2.6222)	(-.1417±.1001)	0.895
Proteus vulgaris					
12	3.4883	3.4136	(6.8254±10.650)	(-.1743±.5893)	0.777
Pseudomonas aeruginosa					
12	11.588	1.8844	(7.4373±3.3536)	(-.1493±.1280)	0.853
Serratia marcescens					
12	27.099	0.9017	(7.6250±2.3198)	(-.1579±.0885)	0.931
Total enteric count (all six above)					
12	21.552	1.0055	(8.1321±2.4497)	(-.1487±.0935)	0.915

Table A-46

Reduction statistics of enteric bacteria species in algal growth medium. Series VI.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	2.2010	2.7828	(6.9431±1.2267)	(-.0228±.0279)	0.180
Enterobacter aerogenes					
12	0.3045	3.6089	(6.0233±1.3969)	(-.0097±.0317)	0.029
Escherichia coli					
12	2.1764	2.4862	(6.3741±1.1595)	(-.0214±.0263)	0.179
Proteus vulgaris					
12	0.6976	3.7817	(6.1353±1.4299)	(-.0149±.0325)	0.065
Pseudomonas aeruginosa					
12	1.1722	1.7682	(6.9922±.9778)	(-.0133±.0222)	0.105
Serratia marcescens					
12	0.3976	2.880	(7.1737±1.2479)	(-.0098±.0283)	0.038

Table A-47

Reduction statistics of enteric bacteria species in filtrate from *Anabaena cylindrica* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
11	6.4853	0.2025	(7.7104±2.4823)	(-.0230±.0570)	0.866
Enterobacter aerogenes					
12	5.7437	0.4313	(7.4657±3.6224)	(-.0316±.0832)	0.852
Escherichia coli					
12	3.2604	0.2060	(8.0062±2.5035)	(-.0164±.0575)	0.765
Proteus vulgaris					
12	2.5121	0.6346	(7.4696±4.3939)	(-.0253±.1009)	0.715
Pseudomonas aeruginosa					
12	3.7237	0.1872	(7.7903±2.3862)	(-.0167±.0548)	0.788
Serratia marcescens					
12	2.2672	0.1520	(7.8025±2.1506)	(-.0118±.0494)	0.694

Table A-48

Reduction statistics of enteric bacteria species in filtrate from *Anacystis nidulans* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	11.972	0.2918	(7.8427±2.9796)	(-.0375±.0684)	0.923
Enterobacter aerogenes					
12	53.426	0.1023	(7.8034±1.7639)	(-.0469±.0405)	0.982
Escherichia coli					
12	4.1309	0.2715	(7.5976±2.8743)	(-.0212±.0660)	0.805
Proteus vulgaris					
12	9.7873	0.6997	(7.3661±4.6137)	(-.0525±.1060)	0.907
Pseudomonsa aeruginosa					
12	5.2270	0.4748	(8.0166±3.8006)	(-.0316±.0873)	0.839
Serratia marcescens					
12	3.0764	0.7320	(7.4349±4.7190)	(-.0301±.1084)	0.754

Table A-49

Reduction statistics of enteric bacteria species in filtrate from *Gloeocapsa alpicola* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	3.2476	0.1193	(8.5132±1.9048)	(-.0395±.0437)	0.970
Enterobacter aerogenes					
12	1016.5	0.0087	(9.2218± .5138)	(-.0596±.0118)	0.999
Escherichia coli					
12	2537.2	0.0042	(8.6674± .3579)	(-.0656±.0082)	0.999
Proteus vulgaris					
12	93.455	0.0594	(9.1244±1.3448)	(-.0473±.0309)	0.989
Pseudomonsa aeruginosa					
14	7.3096	0.1320	(8.6017±2.0040)	(-.0197±.0460)	0.879
Serratia marcescens					
12	0.3661	2.9428	(7.7528±9.4620)	(-.0208±.2174)	0.268

Table A-50

Reduction statistics of enteric bacteria species in filtrate from *Nostoc muscorum* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
14	6.2929	2.9351	(7.5992±9.4495)	(-.0862±.2171)	0.863
Enterobacter aerogenes					
11	29.8502	0.8679	(8.2287±5.1385)	(-.1021±.1180)	0.967
Escherichia coli					
12	22.339	0.8631	(8.0406±5.1242)	(-.0881±.1177)	0.957
Proteus vulgaris					
12	6768.7	0.0038	(8.2417± .3411)	(-.1021±.0078)	0.999
Pseudomonsa aeruginosa					
12	8.9025	1.1393	(7.5259±5.8874)	(-.0639±.1352)	0.899
Serratia marcescens					
12	15.896	0.5653	(7.6208±4.1472)	(-.602 ±.0953)	0.941

Table A-51

Reduction statistics of enteric bacteria species in filtrate from *Oscillatoria chalybia* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	18.221	1.2523	(8.0115±6.4142)	(-.0996±.1473)	0.948
Enterobacter aerogenes					
12	19.231	1.1985	(7.4310±6.0384)	(-.0963±.1387)	0.951
Escherichia coli					
12	8.7375	2.9108	(7.7619±9.4103)	(-.1012±.2162)	0.897
Proteus vulgaris					
12	13.628	1.6409	(7.2278±7.0653)	(-.0949±.1623)	0.932
Pseudomonsa aeruginosa					
12	54.986	0.3240	(7.3250±3.1396)	(0.0847±.0721)	0.982
Serratia marcescens					
12	5.2723	3.0540	(7.7516±9.6390)	(-.0805±.2214)	0.840

Table A-52

Reduction statistics of enteric bacteria species in filtrate from *Oscillatoria formosa* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	2.3677	6.0714	(7.1292±13.5907)	(-.0761±.3122)	0.703
Enterobacter aerogenes					
12	7.9584	1.6716	(8.1111± 7.1312)	(-.0732±.1638)	0.888
Escherichia coli					
12	0.7880	0.5797	(3.1487± 4.1997)	(.0136±.0965)	0.441
Proteus vulgaris					
12	5.0901	2.2407	(7.5433± 8.2564)	(-.0678±.1897)	0.836
Pseudomonsa aeruginosa					
12	24.278	0.3300	(8.4717± 3.1687)	(-.0568±.0728)	0.960
Serratia marcescens					
12	4.2481	2.6330	(8.1498± 8.9501)	(-.0671±.2056)	0.809

Table A-53

Reduction statistics of enteric bacteria species in filtrate from *Phormidium faveolanum* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	6.9894	1.7256	(7.4942±7.2456)	(-.0697±.1664)	0.875
Enterobacter aerogenes					
14	14.285	1.2290	(8.1289±6.1147)	(-.0841±.1405)	0.934
Escherichia coli					
14	2.4922	4.1571	(7.7636±11.2459)	(-.0646±.2584)	0.714
Proteus vulgaris					
11	28.032	0.3666	(7.7698±2.2297)	(-.0643±.0767)	0.965
Pseudomonsa aeruginosa					
13	8.2261	1.4944	(7.6431±6.7427)	(-.0704±.1549)	0.892
Serratia marcescens					
13	14.772	1.1057	(8.6154±5.7998)	(-.0811±.1332)	0.936

Table A-54

Reduction statistics of enteric bacteria species in filtrate from *Ankistrodesmus braunii* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	3.0478	4.8141	(7.7835±12.1020)	(-.0769±.2780)	0.753
Enterobacter aerogenes					
12	12.906	1.7917	(7.9340±7.3829)	(-.0965±.1696)	0.928
Escherichia coli					
14	1342.78	0.0050	(7.3121± .3902)	(-.0520±.0090)	0.999
Proteus vulgaris					
14	93.518	0.2169	(7.3531±2.5691)	(-.0904±.0590)	0.989
Pseudomonsa aeruginosa					
14	55.977	0.3512	(7.9651±3.2685)	(-.0890±.0751)	0.982
Serratia marcescens					
12	8.6920	0.8912	(7.2218±5.2069)	(-.0558±.1196)	0.897

Table A-55

Reduction statistics of enteric bacteria species in filtrate from Chlorella pyrenoidosa at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	5.4223	3.9217	(7.4986±10.9228)	(-.0925±.2509)	0.844
Enterobacter aerogenes					
12	6.9282	2.5946	(7.5341±8.8844)	(-.0851±.2041)	0.874
Escherichia coli					
12	1.8148	6.8664	(7.2231±14.4531)	(-.0708±.3320)	0.645
Proteus vulgaris					
12	5.1004	3.3545	(7.3782±10.1021)	(-.0830±.2321)	0.836
Pseudomonas aeruginosa					
12	0.8458	8.8804	(7.3886±16.4367)	(-.0550±.3776)	0.458
Serratia marcescens					
12	1.4244	8.2050	(7.5455±15.7993)	(-.0686±.3630)	0.587

Table A-56

Reduction statistics of enteric bacteria species in filtrate from Chlorella vulgaris at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
14	8.1485	2.2109	(7.0826±8.2012)	(-.0852±.1884)	0.891
Enterobacter aerogenes					
14	11.8899	0.8985	(5.9278±5.2283)	(-.0656±.1201)	0.922
Escherichia coli					
14	6.2986	1.2075	(7.0093±6.0610)	(-.0553±.1392)	0.863
Proteus vulgaris					
14	14.888	0.9550	(6.5244±5.3902)	(-.0757±.1238)	0.937
Pseudomonas aeruginosa					
14	3.6679	3.1607	(7.2067±9.8060)	(-.0683±.2253)	0.786
Serratia marcescens					
14	2.6636	3.8659	(6.5441±10.8448)	(-.0644±.2491)	0.727

Table A-57

Reduction statistics of enteric bacteria species in filtrate from *Scenedesmus obliquus* at mid-log growth phase. Series VII.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
14	91.4609	0.0285	(8.8573±.9313)	(-.0324±.0214)	0.989
Enterobacter aerogenes					
14	109.62	0.0585	(8.9597±1.3346)	(-.0508±.0307)	0.991
Escherichia coli					
14	51.6465	0.0773	(8.4025±1.5335)	(-.0401±.0352)	0.981
Proteus vulgaris					
14	1.3987	0.4076	(8.2736±3.5216)	(-.0151±.0809)	0.583
Pseudomonas aeruginosa					
14	16.5995	0.4526	(8.0305±3.7108)	(-.0550±.0852)	0.943
Serratia marcescens					
14	5.1187	5.1345	(8.3673±3.9523)	(-.0325±.0908)	0.836

Table A-58

Reduction statistics of pathogenic bacteria species with *Anabaena cylindrica*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	222.0690	.1189	(6.0783±.3755)	(-.0751±.0098)	.9737
Salmonella typhosa					
8	11.0652	1.5269	(6.6067±1.3458)	(-.0601±.0351)	.6484
Shigella paradysenteriae					
8	95.2742	.2458	(6.8035±.5399)	(-.0707±.0141)	.9408
Shigella dysenteriae					
8	106.4579	.2418	(6.7365±.5356)	(-.0742±.0139)	.9466
Vibrio comma					
8	52.2653	.2336	(5.1157±.5265)	(-.0511±.0137)	.8970

Table A-59

Reduction statistics of pathogenic bacteria species with *Anacystis nidulans*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	88.4104	.3737	(7.3962±.6658)	(-.0840±.0174)	.9364
Salmonella typhosa					
8	87.6496	.3064	(6.3929±.6028)	(-.0758±.0157)	.9359
Shigella paradysenteriae					
8	36.2172	1.2425	(6.6402±1.2140)	(-.0981±.0317)	.8579
Shigella dysenteriae					
7	146.9488	.2260	(6.8716±.5993)	(-.1249±.0208)	.9671
Vibrio comma					
7	66.3376	.3186	(5.2523±.7116)	(-.0997±.0247)	.9299

Table A-60

Reduction statistics of pathogenic bacteria species with *Gloeocapsa alpicola*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	10.0872	2.0012	(7.5863±1.5407)	(-.0657±.0401)	.6270
Salmonella typhosa					
8	57.9672	.2999	(5.6856±.5965)	(-.0609±.0156)	.9062
Shigella paradysenteriae					
8	341.4663	.0948	(6.5899±.3353)	(-.0832±.0087)	.9827
Shigella dysenteriae					
8	250.8821	.1036	(5.8648±.3505)	(-.0745±.0091)	.9766
Vibrio comma					
8	19.9350	1.3396	(6.2735±1.2606)	(-.0755±.0329)	.7687

Table A-61

Reduction statistics of pathogenic bacteria species with *Oscillatoria chalybia*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	14.8942	1.1813	(6.6241±1.1837)	(-.0613±.0309)	.7128
Salmonella typhosa					
8	11.2542	1.1372	(5.5874±1.1614)	(-.0523±.0303)	.6523
Shigella paradysenteriae					
8	15.7010	1.3518	(6.3148±1.2663)	(-.0673±.0330)	.7235
Shigella dysenteriae					
8	61.3114	.3928	(6.2078±.6826)	(-.0717±.0178)	.9109
Vibrio comma					
8	228.8732	.0834	(5.1152±.3146)	(-.0639±.0082)	.9745

Table A-62

Reduction statistics of pathogenic bacteria species with *Oscillatoria formosa*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	67.3622	.4893	(6.2018±.7618)	(-.0839±.0199)	.9182
Salmonella typhosa					
8	206.1572	.1061	(5.2634±.3547)	(-.0684±.0093)	.9717
Shigella paradysenteriae					
8	291.2393	.0623	(5.3232±.2718)	(-.0622±.0071)	.9798
Shigella dysenteriae					
8	37.9589	.5839	(5.0350±.8323)	(-.0688±.0217)	.8635
Vibrio comma					
8	84.3495	.2766	(5.0534±.5728)	(-.0706±.0149)	.9336

Table A-63

Reduction statistics of pathogenic bacteria species with *Phormidium faveolarum*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	209.2254	.1396	(6.3641±.4069)	(-.0790±.0106)	.9721
Salmonella typhosa					
8	35.9760	.4714	(4.8470±.7478)	(-.0602±.0195)	.8571
Shigella paradysenteriae					
8	72.4309	.4044	(5.5891±.6926)	(-.0791±.0181)	.9235
Shigella dysenteriae					
8	48.7067	.4307	(5.1532±.7147)	(-.0670±.0186)	.8903
Vibrio comma					
8	59.2853	.3413	(4.6406±.6363)	(-.0658±.0166)	.9081

Table A-64

Reduction statistics of enteric bacteria in algal growth medium under anaerobic conditions.

N	S_H^2/S_r^2	S_r^2	b	k	R
Alcaligenes faecalis					
12	1.089	.7497	(4.2661±1.0626)	(-.0131±.0253)	.179
Enterobacter aerogenes					
12	16.338	.3617	(6.0776±.7381)	(-.0352±.0175)	.7657
Escherichia coli					
12	189.839	.0604	(6.8443±.3016)	(-.0490±.0072)	.9743
Proteus vulgaris					
12	.738	1.1097	(4.7693±1.2928)	(-.0131±.0307)	.1287
Pseudomonas aeruginosa					
12	6.010	.7858	(5.7946±1.0879)	(-.0315±.0259)	.5459
Serratia marcescens					
12	111.430	.1359	(7.5824±.4524)	(-.0563±.0108)	.9571

Table A-65

Reduction statistics of pathogenic bacteria species in algal growth medium under anaerobic conditions.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
12	2.0283	1.0080	(5.2320±1.2322)	(-.0207±.0293)	.2886
Salmonella typhosa					
12	4.2477	.8056	(4.6221±1.1016)	(-.0268±.0262)	.4593
Shigella dysenteriae					
12	46.3827	.2490	(6.5081±.6124)	(-.0492±.0146)	.9027
Shigella paradysenteriae					
12	1.2762	1.4087	(4.2391±1.4567)	(-.0194±.0346)	.2033
Vibrio comma					
12	2.4089	.5228	(4.1682±.8874)	(-.0162±.0211)	.3251

Table A-66

Reduction statistics of pathogenic bacteria in presence of culture of four green algae species. Bacteria added to algae when in their mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
12	35.0416	.3373	(5.0683±1.0587)	(-.1553±.0617)	.9211
Salmonella typhosa					
12	37.5767	.3051	(5.8014±.8522)	(-.1156±.0402)	.9038
Shigella paradysenteriae					
12	46.6440	.3042	(5.7432±1.0055)	(-.1702±.0586)	.9396
Shigella dysenteriae					
12	48.0406	.1846	(5.4882±.7831)	(-.1345±.0457)	.9412
Vibrio comma					
12	41.3008	.2528	(5.0728±.9165)	(-.1460±.0535)	.9323

Table A-67

Reduction statistics of pathogenic bacteria in presence of culture of six blue-green algae species. Bacteria added to algae when in their mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
12	34.9980	.3504	(5.6754±.7463)	(-.0759±.0259)	.87500
Salmonella typhosa					
12	40.8310	.3134	(4.9758±.7058)	(-.0775±.0244)	.89099
Shigella paradysenteriae					
12	36.7898	.1841	(4.2882±.5409)	(-.0564±.0187)	.88044
Shigella dysenteriae					
12	12.6987	.6502	(5.8225±1.4382)	(-.1124±.0673)	.76055
Vibrio comma					
12	53.4203	.1396	(4.9777±.5765)	(-.0933±.0272)	.93033

Table A-68

Reduction statistics of pathogenic bacteria species in algal growth medium Controls.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	7.084	2.936	(4.7225±1.6467)	(-.0622±.0454)	.5414
Salmonella typhosa					
8	75.1391	.3737	(5.3975±.6658)	(-.0775±.0174)	.92605
Shigella paradysenteriae					
8	26.9832	.9442	(5.0349±1.0583)	(-.0738±.0276)	.8181
Shigella dysenteriae					
8	30.5534	.8124	(5.0675±.9817)	(-.0728±.0256)	.8359
Vibrio comma					
8	35.4679	.5158	(4.3030±.7822)	(-.0625±.0204)	.8553

Table A-69

Reduction statistics of pathogenic bacteria species with *Ankistrodesmus braunii*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	53.4130	.4618	(5.2557±.7401)	(-.0726±.0193)	.8990
Salmonella typhosa					
8	50.0659	.4051	(4.5382±.6932)	(-.0658±.0181)	.8930
Shigella paradysenteriae					
8	32.6928	.7621	(4.9435±.9508)	(-.0730±.0250)	.8449
Shigella dysenteriae					
8	25.4007	.9202	(4.7347±1.0448)	(-.0707±.0272)	.8089
Vibrio comma					
8	62.7151	.2627	(4.1532±.5582)	(-.0593±.0146)	.9127

Table A-70

Reduction statistics of pathogenic bacteria species with *Chlorella pyrenoidosa*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
6	16.7589	1.0565	(5.0966±1.3713)	(-.0950±.0495)	.8073
Salmonella typhosa					
6	29.6214	.6437	(5.4513±1.0704)	(-.0986±.0386)	.8810
Shigella paradysenteriae					
6	66.4033	.2931	(5.1800±.7223)	(-.0996±.0261)	.9432
Shigella dysenteriae					
6	11.8522	1.1679	(4.4327±1.4418)	(-.0840±.0520)	.7477
Vibrio comma					
6	17.2831	.6330	(4.3459±1.0614)	(-.0747±.0383)	.8121

Table A-71

Reduction statistics of pathogenic bacteria species with *Chlorella vulgaris*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
7	16.3117	1.2757	(4.6525±1.2950)	(-.0669±.0334)	.7654
Salmonella typhosa					
7	13.2037	1.1533	(4.5957±1.2314)	(-.0572±.0317)	.7253
Shigella paradysenteriae					
7	15.7464	1.1842	(4.1637±1.2477)	(-.0633±.0322)	.7590
Shigella dysenteriae					
7	29.2807	.7786	(4.7370±1.0117)	(-.0700±.0261)	.8541
Vibrio comma					
7	10.9643	.9153	(3.5452±1.0970)	(-.0465±.0283)	.6868

Table A-72

Reduction statistics of pathogenic bacteria species with *Scenedesmus obliquus*. Bacteria added to algae in mid-log growth phase.

N	S_H^2/S_r^2	S_r^2	b	k	R
Salmonella paratyphi					
8	34.3155	.9453	(6.2250±1.0590)	(-.0833±.0276)	.8512
Salmonella typhosa					
7	13.7076	.7617	(4.5996±1.1003)	(-.0700±.0381)	.7327
Shigella paradysenteriae					
7	27.1390	.8834	(6.1021±1.1849)	(-.1061±.0411)	.8444
Shigella dysenteriae					
7	18.2956	1.0167	(5.3369±1.2712)	(-.0935±.0440)	.7854
Vibrio comma					
7	13.4125	1.2051	(4.9428±1.3843)	(-.0872±.0480)	.7285

APPENDIX B

BACTERIOLOGICAL DATA FROM LABORATORY AND FIELD WASTE STABILIZATION POND STUDIES

Table B-1. Total Bacteria Densities In Laboratory Scale Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
7- 3-69	6.72673	6.66346	5.97081	5.54407	5.10380	3.32593	5.65992	4.88081	6.41119	5.53593	5.22011	4.69940
7- 7-69	6.46613	5.97772*	5.50106*	5.08991	6.12222	4.92942*	6.11227*	5.68574*	4.00000*	5.23045	6.65002	6.07188*
7- 9-69	7.05757	6.99717	6.72148	6.90227	5.96190	6.81023	6.58743	5.99388	5.48714	5.64444	6.13928	6.28780
7-11-69	7.34782	7.07372	6.41330	5.78426	5.98520	6.60152	5.69174	6.23553	5.43377	5.58546	5.62273	5.94052
7-14-69	7.19576	6.10380	4.65369	4.79727	5.50718	5.04115	5.47857	4.64836	5.32919	5.49406	5.24613	5.20276
7-16-69	7.49066	6.79239	5.01072	5.09777	5.44248	4.59660	5.68679	4.97058	6.02794	5.08955		3.87506
7-18-69	8.52022	6.79379	4.95425	4.91803	5.48572	4.97405	6.04139	4.53782	5.62014	6.38382	5.21617	4.58433
7-23-69	7.92505	7.47276	6.50827		6.20352	5.34193	5.79344	5.07555	5.67486	6.44739	5.71012	4.38075
7-25-69	7.79449	6.26834	5.44871	5.19728	5.04922	4.34133	5.94374	5.98989	5.42488	5.31597	5.77815	4.09377
7-29-69	7.12385	6.28948*	5.01589*	4.75587	5.13672	4.33445*	5.22272*	5.48180*	5.51455*	5.79623	5.43933	4.98453*

*Inoculation with laboratory cultures: 7-7 and 7-29 with E. c., Pseud., and Serr.

Table B-2. Total Bacteria Densities In Laboratory Scale Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
8- 4-69	7.32531	6.26411	5.12988	4.69152	5.30211	6.19209	5.13751	5.63220	5.45255	5.85643	4.94349	4.96848
8- 6-69	7.66229	5.62685	4.32736	4.05115	3.97428	4.98080	5.56732	5.33244	5.08279	4.96497	4.75397	4.51455
8- 8-69	6.78013	4.74819	6.95624	6.81471	7.21885	7.60590	8.55781	8.46310	7.60487	8.38462	7.65715	7.34922
8-11-69	6.74036	5.67669	4.19033	4.31175	4.61805	4.07278	5.67669	5.04139	5.29003	5.37107	5.35218	4.75967
8-13-69	7.04630	6.17099	5.29667	4.63347	5.40184	4.15381	5.88804	5.09552	5.08458	5.13988	5.26717	5.09691
8-15-69	6.96379	6.24304	5.36549	5.23553	5.02531	4.48714	6.29831	5.16732	5.53782	5.73139	4.93197	5.70948
8-18-69	7.42243	5.47712	4.75397	4.45864	5.12711	4.48572	6.13513	4.92428	5.39226	5.46613	4.85126	5.13830
8-20-69	7.12710	6.02531	3.98677	4.81624	4.49136	4.29994	6.30428	4.07188	5.31175	5.41497	5.86332	5.09342
8-22-69	7.51851	6.36680	5.20412	5.94694		4.61278	6.69329	5.49136	5.70372	5.72937	5.93952	5.54064
8-26-69	6.87216	5.30103	4.16137	4.52504	4.43537	4.11227	4.79588	4.70757	4.57113	4.68350	5.02119	4.59934

Table B-3. Total Coliform Bacferia Densities In Labor tory Scale Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	7	#8	#9	#10	#11
7- 3-69		4.88930	4.39794		2.08279	0.74036	3.71642	0. 9794	3.87506	3.54407	2.00000	
7- 7-69	5.14613	3.92942*	*			*	4.39902*	*	2.00000*	3.97772	2.30103	*
7- 9-69	4.69907	5.07918	4.55630	5.59106	4.30103	4.90714	3.96614	4.6 7482			4.00000	3.26682
7-11-69	5.60746	5.00000	5.36173	4.50515	5.06070	4.74036	4.30103	4.6 206	4.14613	4.38021	2.14922	
7-14-69		5.07918	3.13830	3.62839	3.90309	3.00000	3.00000	3.7 815	3.15381	3.12222	3.56526	4.23045
7-16-69	6.81258	6.32222			4.13033							
7-18-69	5.87216	6.43537	3.79588	2.81291	3.79588		4.63599	3.7 967	4.76343	4.76080	4.17609	1.57978
7-23-69	6.66039	4.70329	0.97772	0.17609	0.17609	4.31175	3.72632	4.2 539	3.27875	2.87040	4.49693	2.13033
7-25-69	5.62839	4.73679					3.87535		2.39750	2.95904	2.51587	
7-29-69	6.01807	4.28443*	*			*	2.95036*	*	2.72815*	2.79571	2.63246	*

*Inoculation with laboratory cultures: 7-7 and 7-29 with E. c., Pseud., and Serr.

Table B-4. Total Coliform Bacteria Densities In Laboratory Scale Wastes Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
8- 4-69	5.19866	3.40449	1.72222	0.54407		3.00432	2.02632	2.20140	2.05881	1.80787	2.09078	1.74624
8- 6-69	5.85126		1.07918	0.84510	0.30103	1.97543		1.23045		1.79934	0.69897	0.90309
8- 8-69	5.75587	4.06633	0.30103	0.30103		1.02110	2.95424	1.06446	3.61278	0.95424	1.92428	
8-11-69	5.71600	3.81291	1.49136	2.32222		0.81291	3.17609	0.30103	3.06446	3.27875	3.23045	2.30103
8-13-69	5.78176	4.39794				1.25527	1.99123	1.51188	3.51851	3.50515	3.14613	1.76343
8-15-69	5.57978	4.65321	0.77815	2.04139		3.30103	4.65321	3.17609	3.69897	4.17609	4.53148	4.43136
8-18-69	5.08279	4.37566		0.00000	0.00000	0.90309	3.29003	1.72428	2.38021	3.34242	3.27875	2.40867
8-20-69	6.10551	4.51521		0.00000		1.71391	2.95424		3.14613	3.27875	3.11394	0.17609
8-22-69	5.86332	3.84510	0.17609	0.00000		1.71181	2.52114	0.00000	3.17609	3.14613	3.32736	0.30103
8-26-69	5.72222	3.84510		0.00000		2.04238	2.49554	0.17609	2.77815	2.51055	2.13672	1.07918

Table B-5. Escherichia coli Densities In Laboratory Scale Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
7- 3-69		4.54407										
7- 7-69	4.74036	3.74036*	*			*	3.00496*	*	*		3.00000	*
7- 9-69	4.69954	5.07918	4.55630	5.59660	4.30103	4.90687	3.96614	3.67685			4.00000	3.03080
7-11-69		4.69397	5.36173	4.38021	4.84510	5.74036	4.30103	4.39794				1.68124
7-14-69		4.95424	3.13033	3.62839	3.90309	3.00000	3.00000	3.77815	3.15381	3.12222	3.56526	4.07918
7-16-69	5.49381	6.29447	3.00000		3.17609	4.30103	4.02119	3.47712	3.17609	3.60206	3.90309	3.00000
7-18-69	5.27875	5.06004					4.11394	2.74036	3.84510	4.04115	3.77815	1.54407
7-23-69	6.49206	4.86332	0.00000			3.60206	3.21748	2.92942	2.03443	0.00000	4.44560	1.11394
7-25-69	5.06070	4.11394							1.14613	2.81291	2.60206	
7-29-69	5.41497	3.62325*	*			*	2.41664*	*	3.17609*	1.89487	2.02531	*

*Inoculation with laboratory cultures: 7-7 and 7-29 with E. c., Pseud., and Serr.

Table B-6. Escherichia coli Densities In Laboratory Scale Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
8- 4-69	4.95785	2.20276	1.72222	0.54407		3.00453	1.90309	2.12385	2.02531	1.14613	2.09078	1.74624
8- 6-69	4.37107		0.90309	0.60206		1.97543		0.37506		1.30103		0.90309
8- 8-69	4.14613	3.27875	0.00000			0.69897	2.07918	0.84510	3.00000		1.11394	
8-11-69	3.65321					0.30103	3.07918	0.00000	3.84510	3.92942	2.69897	2.30103
8-13-69	5.21748	3.81291					1.30103	0.00000	2.60206	2.54407	2.30103	
8-15-69	4.92942	4.00000				0.54407	0.47712			2.00000	2.77815	
8-18-69	5.55023	3.54407		0.00000		0.84510	2.69897	1.70969	2.92942	2.84510	2.95424	0.69897
8-20-69	5.27875	4.14613				1.70969	2.54407		2.47712	2.65321	2.47712	0.00000
8-22-69	5.26717	3.54407		0.00000		1.71181	1.73640		2.30103	2.79588	2.81291	0.00000
8-26-69	5.00432	3.39794				2.03941	0.39794		2.30103	1.73838	1.71181	

Table B-7. *Pseudomonas aeruginosa* Densities In Laboratory Scale Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
7- 3-69			4.30103			0.00000			4.00000	3.77815	3.74036	
7- 7-69		*	*			*	*	*	*			*
7- 9-69			5.97772	5.97772	5.17609	5.34242		3.00000				
7-11-69		4.00000	5.30103		3.87506	4.92942	3.00000	3.81291			4.00000	3.00000
7-14-69										2.00000		
7-16-69												
7-18-69												
7-23-69								4.00000				
7-25-69												
7-29-69		*	*			*	*	*	*			*

*Inoculation with laboratory cultures: 7-7 and 7-29 with E. c., Pseud., and Serr.

Table B-8. *Pseudomonas aeruginosa* Densities In Laboratory Scale Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station--											
	Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
8- 4-69			4.17609				3.30103	3.69897		3.00000	3.47712	
8- 6-69			3.00000						3.00000			
8- 8-69												
8-11-69												
8-13-69												
8-15-69					3.00000					3.00000		3.84510
8-18-69											3.00000	3.00000
8-20-69												3.00000
8-22-69			3.00000	3.77815		2.30103				4.74036		
8-26-69	5.30103		3.00000	3.74036	2.90309	2.79588		3.00000	3.00000	3.39794		

Table B-9. Serratia marcescens Densities In Laboratory Scale Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
7- 3-69												
7- 7-69		*	*			*	*	*	*			*
7- 9-69			5.64836	5.53782	4.73038	4.74036	3.77815	3.90309		3.77815		
7-11-69			4.75967	3.47712							4.00000	
7-14-69			2.81291	2.92942								4.30103
7-16-69												
7-18-69				2.00000					4.90309			
7-23-69												
7-25-69												
7-29-69		*	4.12222*	4.24304	3.95424	*	3.00000*	4.30103*	*		4.00000	*

*Inoculation with laboratory cultures: 7-7 and 7-29 with E. c., Pseud., and Serr.

Table B-10. *Serratia marcescens* Densities In Laboratory Scale Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station--											
	Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
8- 4-69			4.30103	4.16137		3.77815		4.19033		3.00000	3.39794	
8- 6-69			4.19033	3.30103								
8- 8-69											4.74036	
8-11-69												
8-13-69												
8-15-69							3.00000					
8-18-69	5.00000											
8-20-69												
8-22-69												
8-26-69												

Table B-11. Chromagen Densities In Laboratory Scale Waste Stabilization Ponds, As $\mathrm{Log}_{10}/\mathrm{ml}$.

	Sample Station--											
Date	Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
7- 3-69		5.36642	5.29003	4.74036	4.38021		4.70757	4.09691	5.62325	3.95424	4.52504	4.13033
7- 7-69						4.00000					5.65992	
7- 9-69	5.07004	5.45484					6.02531	5.74036	4.86332	5.07555	5.74710	
7-11-69	5.84510	5.32222	5.54407	4.64836	5.37475	6.26245	5.01599	5.91566	4.74429	5.03743	5.07918	5.63548
7-14-69			3.87506	3.74036	4.06070		3.74036	4.00000	2.81291	3.66978	4.10551	4.30103
7-16-69			3.84510	3.77815	4.27875		4.77761	3.79588	3.19033	3.30125		
7-18-69												
7-23-69	6.06070	5.47712	4.52114	3.77815	5.35781	4.35784	5.09342	4.07918	4.51851	6.00065	4.91116	2.55023
7-25-69	6.63849		3.47712	3.00000			4.41497	4.34242	4.60206	3.47712	4.77815	0.81291
7-29-69				3.47712	4.27875	2.69897	3.87506	4.46613	4.63347	4.77452		3.19033

Table B-12. Chromagen Densities In Laboratory Scale Waste Stabilization Ponds, As $\mathrm{Log}_{10}/\mathrm{ml}$.

Date	Sample Station-- Raw	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11
8- 4-69	6.47712	4.94448	4.07004	3.86923	5.11394	4.7493	4.61542	5.17245	5.37566	5.85431	3.55023	4.73038
8- 6-69	5.00000				3.07004		3.74036	4.00000		3.00000		3.84510
8- 8-69	5.69897		6.95424	6.81291	7.21748	7.60206	8.55630	8.46240	7.60206	8.38021	7.65321	7.34242
8-11-69			3.47712		3.00000	2.91645	2.69897	4.95424	4.30103	4.30103	4.47712	4.09691
8-13-69			2.47712		5.10037	3.94511	5.67210	5.06070	4.00000	3.47712		5.01284
8-15-69			4.81291	4.03141	4.30103	3.85278	5.43933	4.62325	5.09691	5.14998	4.41498	5.30103
8-18-69			4.04139	3.74036	4.04139				4.72428			
8-20-69												
8-22-69	6.17609		4.61805	5.19033		3.75967	5.04139	4.54407	5.08991	5.24304	5.46240	4.93450
8-26-69												

Table B-13. Total Bacteria Densities In Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
6- 4-69	6.34242	6.35218		6.51455		4.98677	6.15534		5.66276			
6- 5-69	6.44716	6.57978*	6.30103	5.92428*	5.32222	5.07918	5.66276	5.60206				
6- 6-69	6.73560	6.69897	7.00000	6.14922	5.43136	5.69020	6.28780	5.69897	5.56820			
6- 9-69	6.70842	6.02119	6.67394	6.20140	6.58883	5.74036	6.65610	5.19866	4.84510			
6-11-69	6.70415	6.38202	6.35025	4.81954	5.55630	4.56820	5.42975	5.39794	4.77815	5.80482	5.79029	
6-13-69	6.69984	6.58659	6.28556	3.00000	5.29667	4.79934	5.45025	5.41497	4.94448	6.49136	6.25285	
6-16-69	6.59550	5.98677	5.95904	4.74819*	5.67025	4.74036	5.28330	6.34830	4.44716	4.44716	5.29003	5.34242
6-18-69	6.96100	5.72673	5.90472	5.06819	5.67302	5.24304	5.17319	5.29885	3.60206	5.17898	4.79239	3.30103
6-20-69		6.46240	6.29885	4.60206	4.47712	4.27875	*		3.00000			3.77815
6-23-69		6.82217	6.48430			4.57978	3.00000		4.85126	*		3.60206
6-25-69						3.95424			3.04139			3.87506
6-27-69	6.69108	6.65706	6.64246	3.14613	4.46850	3.20412	3.57978	3.50515	3.20412	5.46538	5.65992	3.89763
6-30-69	7.03262	6.35458	6.34044	6.96656	4.38758	3.25696	4.57980	5.70672	3.66783	5.65300	5.78247	4.48195

*Inoculation with laboratory cultures: (6-5; E. c., Pseud.) (6-16; E. c., Pseud., Serr.) (6-19; shown as 6-20; E. C., Pseud., Serr.) (6-23; E. c., Pseud., Serr.)

Table B-14. Total Bacteria Densities In Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
7- 2-69	6.73878	6.98644	6.93865	5.37220	5.48853	4.10072	5.14426	5.23259	5.18611	5.72835	5.76530	5.30604
7- 7-69	7.29612	7.35362	7.19590	5.43616	5.58092	3.58192	5.21484	4.98227	4.74819	5.22660	5.61013	5.07004
7- 9-69	6.86629	6.36605	7.03993	3.87344	5.42922	5.47787	5.17342	5.40747	4.87484	6.27140	6.63624	5.74321
7-11-69	7.34587	7.27646	7.53656	5.36577	6.09412	5.28780	6.64147	6.22154	5.88053	6.21163	6.37658	5.85187
7-16-69	7.19576	7.37767	7.10806	5.59106	5.78319	4.16443	5.51521	5.38471	4.52827	6.44091	6.38292	5.96308
7-18-69	7.49066	6.60016	6.03523	6.63829	6.48053	6.18064	6.59555	6.35005	5.33011	6.82086	6.62428	6.62926
7-21-69	8.52088	7.96497	8.25600	5.77706	6.19089	4.86540	6.33163	6.18227	5.25139	6.38739*	7.22789	6.43553
7-23-69	7.92505	7.19033	7.34635	5.53013	6.73632	6.02735	6.50853	6.57119	5.79955	7.12548	6.93717	6.86608
7-25-69	7.79449	6.82445	7.19451	7.28319	7.17713	6.26174	6.28171	6.50127	5.44754	6.72016	7.13815	5.39094
7-29-69	7.12385	7.27646	6.56820	5.20548	5.61262	4.15987	5.29115*	5.36949	4.24055	6.08814	6.14768	5.24920
7-31-69	7.22272	6.92298	7.14301	5.77379	5.52504	4.45102	5.33746	5.26600	2.94201	6.02325	5.56229	4.92169

*Inoculation with laboratory cultures: 7-21 and 7-29 with E. c., Pseud., and Serr.

Table B-15. Total Bacteria Densities In Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
8- 4-69	7.33244	7.24981	6.98000	4.67210	6.33496	5.63246	5.62480	5.56526	5.04336	6.62660	5.63849	4.97313
8- 6-69	7.66229	6.93069	6.99344	6.12215	5.99100	5.55991	5.58574	5.79934	5.37794	5.55961	5.92634	4.98453
8- 8-69	7.05207	7.20880	7.29170	6.40140	6.63849	5.49136	7.20412	6.44770	5.93197	6.46117	6.35338	5.66229
8-13-69	7.04630	7.08636	7.08189	6.88550	6.82102	4.96379	7.05300	6.61316	5.78645	6.34586	6.25467	5.15076
8-15-69	6.96379	7.22789	7.48996	7.28499	7.14613		7.38828	6.84510	5.93827	6.71904	6.66745	6.12548
8-18-69	7.42243	6.91116	7.29447	6.80702	6.87938	6.13909	7.15503	6.73739	6.24748	6.55023	6.87795	5.83727
8-20-69	7.12710	7.22531	7.26186	6.46165	6.71391	6.37475	6.81067	5.79831	5.64738	5.93827	6.02531	5.60959
8-22-69	7.51851	7.29115	7.29336	7.05018	6.87520	6.89708	7.42854	7.25768	6.47276	6.89070	6.30049	6.56926
8-26-69	6.87216	6.84819	6.84973	6.67440	6.54777	5.72428	6.16584	5.72222	5.59988	5.54407	5.55328	5.31755

Table B-16. Total Coliform Bacteria Densities In Waste Stabilization Ponds, As $\mathrm{Log}_{10}/\mathrm{ml}$.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
6- 4-69		5.69897		5.60206	-		5.47712					
6- 5-69	5.47712	5.30103*	5.00000	4.20412*	4.11394	3.90811	4.54407	4.34242	3.77815			
6- 6-69	5.77815	5.60206	5.47712	4.17609	4.39794	2.80625	5.81889	3.90309				
6- 9-69	5.77670	5.70969	5.83569	5.15836	3.69897	1.74115	4.88081	3.47712				
6-11-69	5.98137	5.00000	4.81954		3.00000		3.92942	4.14613		4.74819	4.57403	
6-13-69	5.77815	6.21484	4.93450		3.17609	2.93450		4.25527		4.83885	4.43136	
6-16-69	5.90309	4.77085	4.66276	2.69897*	4.39794	1.63347	3.84510	3.69897		3.74036	3.87506	2.69897
6-18-69	5.02938	5.17319	5.17609	0.69897	1.86923	1.59106	2.71349	3.19285	0.00000	3.37840	2.00043	1.04139
6-20-69	6.23045	5.04139	5.04139	2.20140	1.64345	1.72428	3.00000*	3.84510		3.84510	3.60206	0.97772
6-23-69	5.75587	4.92942	5.43136	4.90309		1.86332	1.13033	0.60206	4.17609	*	3.00000	
6-25-69	5.80618	4.65321	4.96848	2.40483	0.69897		2.88053	1.32222	1.39794	3.03342	2.53020	0.77815
6-27-69	6.51983	4.77815	4.84510	0.69897	1.25527	1.87216	3.04532	3.18752		2.94349	3.48430	0.77815
6-30-69	5.85733	4.60206	4.77815	0.00000	0.54407	1.92942	1.44716	0.45788		0.54407	2.95665	0.30103

*Inoculation with laboratory cultures: (6-5; E. c., Pseud.) (6-16; E. c., Pseud., Serr.) (6-19; shown as 6-20; E. c., Pseud., Serr.) (6-23; E. c., Pseud., Serr.)

Table B-17. Total Coliform Bacteria Densities In Waste Stabilization Ponds, As $\mathrm{Log}_{10}/\mathrm{ml}$.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
7- 2-69	5.62325	4.30300	5.07004	2.70136	0.02119		2.75797	2.71204	0.47712	3.42704	2.77670	2.70286
7- 7-69	6.12304	5.19728	4.74036	0.69897	2.70265	0.30103	3.49534	3.19770	0.60206	3.49631	2.80058	0.47712
7- 9-69	5.59934	5.77815	5.17609	0.60206	2.70243	0.54407	1.31175	3.84510	1.81291	3.69897	0.84510	1.17609
7-11-69	5.65321	5.27875	4.64098		1.21748	0.74036				2.03141	2.81291	
7-16-69	6.81258	5.24304	4.89900	2.94201	3.20412		4.02119	3.94939	1.00000			
7-18-69	5.87216	5.14922	4.89209	2.84261	3.92686		3.46240	3.55630	1.13033	4.48359	3.86332	3.90300
7-21-69	6.17464	4.96848	4.65801		0.00000		3.83727	3.47349	1.29003	*	3.90982	0.54407
7-23-69	6.66039	5.27068	5.51117	2.02735	2.13354	0.90309	4.01912	4.66978	0.30103	4.09342	3.11394	0.77815
7-25-69	5.62839	4.59660	4.94448	0.00000	2.84510		3.66745	3.74036	0.30103	3.37107	3.89487	1.24304
7-29-69	6.01807	4.29003	4.73838	0.00000	2.00000	0.47712	2.74036*	2.97772		1.90445	3.50174	0.47712
7-31-69	4.89209	5.05500	4.06633	2.00304		0.14613	3.31175	2.90309	0.90309	5.13354	3.19033	1.49831

*Inoculation with laboratory cultures: 7-21 and 7-29 with E. c., Pseud., and Serr.

Table B-18. Total Coliform Densities In Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
8- 4-69	5.19866	4.99782	4.86332	0.30103	2.00000	1.30103	2.47712	2.30103	0.77815	3.13033	3.00000	0.77815
8- 6-69	5.85126	4.71809	4.53148	1.78176	2.87852	0.00000	3.51188	3.27875		2.81291	3.75967	
8- 8-69	5.75587	4.54407	4.92942	1.17609	2.88944	1.00000	3.99123	3.52504	1.00000	2.00647	4.59106	0.30103
8-13-69	5.78176	4.96142	4.69461	1.97658	2.55509	0.47712	4.11561	3.46982	0.90309	3.65321	3.60314	1.17609
8-15-69	5.57978	4.81790	4.88081	3.92169	3.47712	1.55630	3.82607	3.67210	1.72016	3.95424	4.14301	2.15076
8-18-69	6.08279	4.97081	3.72428	3.03141	3.09691	0.60206	3.96379	3.72222	0.65321	3.16137		1.71181
8-20-69	6.10551	4.92686	4.96142	2.15381	3.71809	2.47712	3.84354	3.89625	1.72016	3.63849	3.47712	2.01072
8-22-69	5.86332	4.90037	4.65801	2.00432	1.91116	0.00000	3.84819	3.34242	2.41664	3.61013	3.55630	1.78355
8-26-69	5.72222	5.07225	4.84819	3.13409	3.50379		3.29003	3.68124		3.24304	3.74233	2.50651

Table B-19. Escherichia coli Densities In Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
6- 4-69		5.30103										
6- 5-69	5.00000	*		3.84510*	3.30103		4.04139	3.00000	3.00000			
6- 6-69	5.00000	5.00000	5.00000				4.75587	3.75587				
6- 9-69	5.23553	3.92942	4.06070	3.97772	3.30103		4.42813	2.69897				
6-11-69	4.81954	4.11394	4.46240				3.00000	3.39794		3.54407	3.47712	
6-13-69	5.00000	4.88195	4.34242		2.69897			3.81291		3.39794	3.00000	
6-16-69	5.39794	3.69897	3.39794	*			3.17609	3.17609		3.00000		
6-18-69	4.19033	5.03743	3.17609	0.30103					0.77815	1.46240	0.87506	0.54407
6-20-69	5.97772	4.84510	4.72428	1.89763	2.43457		*	3.60206	0.17609	1.69897*	3.47712	0.47712
6-23-69		4.54407	4.60206	0.69897			0.00000	0.39794	3.39794			
6-25-69	5.34242	4.00000	4.39794	0.60206	0.17609		2.40140	0.00000	0.17609	2.94448	1.54407	0.17609
6-27-69	5.67210	4.74036	4.77815	0.00000	0.47712		2.77085	3.02531		3.35218	2.77815	0.00000
6-30-69	5.58546	4.47712	4.65321	0.00000	0.30103		1.30103	1.54407		0.17609	1.99123	

*Inoculation with laboratory cultures: (6-5; E. c., Pseud.) (6-16; E. c., Pseud., Serr.) (6-19; shown as 6-20; E. c., Pseud., Serr.) (6-23; E. c., Pseud., Serr.)

Table B-20. Escherichia coli Densities In Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
7- 2-69	5.04139	4.17609	4.82930	0.30103	0.00000		1.46240	0.77815		2.89154	2.70906	0.60206
7- 7-69	5.46613	5.06070	4.30103	0.39794	0.47712	0.30103	3.24920	3.24834	0.00000	3.00000	1.60746	0.00000
7- 9-69	5.44248		5.00000			0.54407		3.30103		2.00647		
7-11-69		4.68970	3.30103							2.00647		
7-16-69	5.49381	4.69897	4.57403	2.00967	2.00000		3.63347	3.41078	0.30103	3.59660	3.67669	0.81291
7-18-69	5.27875	4.45102	4.41497	2.57692	2.00000		2.90309	3.04139	0.69897	3.43933	3.24304	
7-21-69	5.54407	4.75587	4.10551		0.00000		3.49136	3.50515	0.00000	*	3.03141	0.30103
7-23-69	6.49206	4.97772	4.58546	2.01599	1.90309	0.00000	3.78176		0.00000	3.65801	3.69897	
7-25-69	5.06070	3.92942	4.39794		2.00000		3.14613	2.90309	0.00000	2.81291	3.14613	0.30103
7-29-69	5.41497	4.13830	4.48430				3.47712*	2.54407		2.30103	2.84510	
7-31-69	4.67210	4.78533	3.66976				3.11394	2.77815		4.66276	2.97772	

*Inoculation with laboratory cultures: 7-21 and 7-29 with E. c., Pseud., and Serr.

Table B-21. *Escherichia coli* Densities In Waste Stabilization Ponds, As Log_{10}/ml.

	Sample Station--											
Date	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
8- 4-69	4.95785	4.66745	4.11394						0.30103	2.65321	2.84510	
8- 6-69	5.37107	4.27875	4.14613		0.47712	0.00000	3.20412	3.00000		2.69897	3.27875	
8- 8-69	5.14613	3.90309	4.25527	0.84510	1.72428	0.30103	3.23045	3.84510	0.30103	2.54407		
8-13-69	5.21748	4.51188	4.06691	1.75967	2.18184		2.92942	2.90309	0.00000	2.77815	2.65321	0.30103
8-15-69	4.92942	4.32736	4.16137	3.86332	2.84510	0.60206	3.20412	3.17609	1.25527	2.90309	2.92942	0.65321
8-18-69	5.55023	4.62066	3.43933	2.84510	2.82930		3.56229	3.33244	0.30103	2.95424		2.00000
8-20-69	5.27875	4.14613	4.20412	1.73640	2.00000		3.30103	2.81291	0.30103	3.06070	3.00000	0.00000
8-22-69	5.26717	4.37107	4.14613	0.17609	1.72428		3.26717	2.94201	1.70329	2.90309	2.88930	1.71809
8-26-69	5.00432	4.30643	4.00432	2.14535	2.49693		3.07918	3.04139	2.35218	2.91645	3.13830	0.77815

Table B-22. Pseudomonas aeruginosa Densities In Waste Stabilization Ponds, As $\mathrm{Log}_{10}/\mathrm{ml}$.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
6- 4-69												
6- 5-69		*		*	3.00000							
6- 6-69	5.00000	3.00000										
6-11-69												
6-13-69			3.00000		3.30103							
6-16-69		3.17609		*				3.77815				
6-18-69							3.00000			3.00000		
6-20-69		5.00000	4.00000				*					
6-23-69	4.81291							2.69906	3.00000	3.00000*	5.87506	
6-25-69				0.00000		0.00000		4.60206				
6-27-69												
6-30-69	5.00000			0.60206	0.30103	0.00000						

*Inoculation with laboratory cultures: (6-5; E. c., Pseud.) (6-16; E. c., Pseud., Serr.) (6-19; shown as 6-20; E. c., Pseud., Serr.) (6-23; E. c., Pseud., Serr.)

Table B-23. *Pseudomonas aeruginosa* Densities In Waste Stabilization Ponds, As Log_{10}/ml.

	Sample Station--											
Date	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
7- 2-69			5.30103									3.00000
7- 7-69												
7- 9-69				0.69897	0.97772			0.00000				1.04139
7-11-69						3.00000						
7-16-69												
7-18-69												
7-21-69										*		
7-23-69												
7-25-69												
7-27-69												
7-29-69							*					

*Inoculation with laboratory cultures: 7-21 and 7-29 with E. c., Pseud., and Serr.

Table B-24. Pseudomonas aeruginosa Densities In Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
8- 4-69												
8- 6-69		6.30103		3.47712	3.00000						3.74036	
8- 8-69			5.00000									
8-13-69	6.00000											
8-15-69		5.00000							3.09691		4.30103	3.17609
8-18-69									4.00000			
8-20-69												
8-22-69												
8-26-69	5.30103			4.00000				3.30103	3.74036			3.00000

Table B-25. Serratia marcescens Densities In Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
6- 4-69												
6- 5-69												
6- 6-69												
6- 9-69												
6-11-69												
6-13-69												
6-16-69												
6-18-69					5.00000						3.00000	
6-20-69										*		
6-23-69										3.60206	6.76716	
6-25-69								4.30103				
6-27-69	5.00000				3.69897		0.30103			0.30103		
6-30-69						2.69914	*		3.00000			

*Inoculation with laboratory cultures: 7-21 and 7-29 with E. c., Pseud., and Serr.

Table B-26. *Serratia marcescens* Densities In Waste Stabilization Ponds, As Log$_{10}$/ml.

	Sample Station--											
Date	#1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
8- 4-69												
8- 6-69					3.00000		3.77815		3.00000		4.17609	3.74036
8- 8-69						3.00000	4.04139	4.00000				
8-13-69												
8-15-69										4.00000	4.77815	
8-18-69										4.00000		4.00000
8-20-69							5.17609			4.00000		
8-22-69												
8-26-69												

Table B-27. Chromagen Densities In Waste Stabilization Ponds, As Log_{10}/ml.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
6-30-69	5.17609	5.15381	5.60206	3.35392	3.00475	3.00303	3.00260	3.60206	1.73239	3.44404	3.47712	3.01452
7- 2-69	5.54407	5.39794	5.39794	4.38021			5.04532	4.93952		3.47712	3.97772	
7- 9-69	5.07004	6.11644	6.19451	2.70372	4.77452	5.01912	4.33415	4.51382	4.37493	5.92428	6.27300	4.20656
7-11-69	5.84510	6.96848	7.28103	5.17026	5.53593	4.87338	6.27875	4.38021	5.92763	5.35698	5.69020	5.26007
7-18-69		6.50515	5.77815	6.23465		5.65562	6.17826	5.92763	4.99454	6.30750	5.72835	5.63949
7-21-69	5.69897	5.60206	6.00000	3.00000				5.39794	4.40824	*		6.05500
7-23-69	6.06070	5.69897	5.90309	6.19866	6.74135	5.69858	6.08027	5.96755	5.40697	6.71809	6.59638	6.50583
7-25-69	6.63849	6.32222	6.63347	6.41747	6.86004	5.93044	5.91619	6.04001	4.83569	6.34044	6.71684	
7-29-69			0.00000	3.74086	2.54407		*	2.17609				4.65321
7-31-69	5.00000				3.00000	2.47712						

*Inoculation with laboratory cultures: 7-21 and 7-29 with E. c., Pseud., and Serr.

Table B-28. Chromagen Densities In Waste Stabilization Ponds, As $\text{Log}_{10}/\text{ml}$.

Date	Sample Station-- #1	#2	#3	#4	#5	#6	#7	#8	#9	#10	#11	#12
8- 4-69	6.47712	6.53148	6.00000	4.91381	6.26717	5.60206	4.21748	5.24428	4.95454	6.61013	5.49136	4.62839
8- 6-69	6.00000	6.27875	6.13830	5.89625	5.82930	5.46761	4.67897	5.33746	5.29885	5.29003	5.25888	4.66511
8- 8-69	5.69897	5.30103		5.96731	6.19728	5.17934		5.39138		5.37143	5.29281	4.81624
8-13-69		6.35218	6.47712	6.84042	6.76343		7.02016	6.54407	5.69897	6.18255	6.10551	
8-15-69		6.35218	6.47712	6.84042	6.76343		7.02016	6.54407	5.69897	6.18255	6.10551	5.54095
8-18-69				6.07004	6.60206	5.57403	7.11394	6.69897	6.19728	6.51188	6.84510	5.60206
8-20-69		6.69897	6.37107	5.69897	6.57978	5.57978	6.70969	5.34242	4.23045	5.69020	5.70757	5.31702
8-22-69	6.17609	6.09691	6.27875	6.45102	6.22337	6.69461	7.07555	6.86629	6.13033	6.51851	5.84819	6.24981
8-26-69												

APPENDIX C

PROGRAM BETA FORMAT

```
                 PROGRAM BETA(INPUT,OUTPUT)
000002           DIMENSION INDEX(14),Y(400),X(20,400),W(400),TABLE(34,3),A(20,20),
                1YX(20),B(20),CDOT(20,20),BDOT(20),CINV(20,20),NSUB(20),QP(20),
                2COVAR(20,20),SU1(20),BSTAR(20),FORM2(5),
                3FORM(7),CN(20,20),C(20,20),VARI(400),YHAT(400)
000002           DIMENSION YHATT(400), YUP(400), YLOW(400), DELTY(400)
000002           DIMENSION  ICELL(10)
000002           COMMON/A/W,N,EP,Y
000002           COMMON/B/TABLE
000002           COMMON/C/N
000002           DATA(TABLE=1.,2.,3.,4.,5.,6.,7.,8.,9.,10.,11.,12.,13.,14.,15.,16.,
                917.,18.,19.,20.,21.,22.,23.,24.,25.,26.,27.,28.,29.,30.,40.,60.,12
                10.,500.,
                56.3138,2.9200,2.3534,2.1318,2.0150,1.9432,1.8946,1.8595,1.8331,
                61.8125,1.7959,1.7823,1.7709,1.7613,1.7530,1.7459,1.7396,1.7341,
                71.7291,1.7247,1.7207,1.7171,1.7139,1.7109,1.7081,1.7056,1.7033,
                81.7011,1.6991,1.6973,1.6839,1.6707,1.6577,1.6449,
                1        12.706,4.3027,3.1825,2.7764,2.5706,2.4469,2.3646,2.3060,
                22.2622,2.2281,2.2010,2.1788,2.1604,2.1448,2.1315,2.1199,2.1098,2.1
                3009,2.0930,2.0860,2.0796,2.0739,2.0687,2.0639,2.0595,2.0555,2.0518
                4,2.0484,2.0452,2.0423,2.0211,2.0003,1.9799,1.9600)
000002         1 FORMAT (I2,A3,5X,A7)
000002        37 FORMAT(3H X(I2,1H)/I25,12X,3F20.10)
000002        38 FORMAT(29H(3H X(I2,17H) AFTER REMOVING I2,14H(2HX(I2,2H) )))
000002        65 FORMAT(5F20.10)
000002        66 FORMAT(9H A-MATRIX /)
000002        67 FORMAT(//29H INVERTED A-MATRIX (C-MATRIX) /)
000002        68 FORMAT(//9H G-MATRIX /)
000002        70 FORMAT(//11H C*A MATRIX/)
000002        75 FORMAT(//30H REGRESSION COEFFICIENTS              /)
000002      1560FORMAT (//85H TOTAL CALCULATED FROM DATA DI6FERS FROM TOTAL CALCUL
                1ATED BY SUMMING COMPONENTS BY                    F20.10//)
000002       600 FORMAT(14A4)
000002       601 FORMAT(2I3)
000002       605 FORMAT(3H B(I2,5H)  = F20.10)
000002       606 FORMAT(  20H DUE TO REGRESSION  I5,12X,3E20.10//15H DEVIATION FROM
                1/11H REGRESSIONI14,12X,2E20.10//6H TOTAL I19,12X,E20.10)
000002       607 FORMAT(I2,2A4)
000002       608 FORMAT(20I2)
000002       614 FORMAT(3H B(I2,5H)  = F20.10,10H    +/-    F20.10)
000002       615 FORMAT(15H VARIANCE ON Y(I3,3H) =F20.10)
000002       616 FORMAT(3HAY(I3,5H)  = F20.10,10H    +/-    F20.10,5X,F20.10,F10.4)
000002      6161 FORMAT (    3HAY(I3,5H)  = F20.10,5X,F20.10,5XF20.10,5XF20.10    )
000002       617 FORMAT(/40H MULTIPLE CORRELATION COEFFICIENT (R) = E20.10//
                1 17H VARIANCE RATIO = E20.10//)
000002       625 FORMAT(6F20.10)
000002       627 FORMAT(5F20.10)
000002       628 FORMAT(//24H CONFIDENCE LIMITS OF B )
000002       651 FORMAT(//17H COVARIANCES ON B//)
000002       652 FORMAT(9H REMOVING )
000002       653 FORMAT(I25,12X,3E20.10)
000002       655 FORMAT(3H X(,I2,10H) REMOVED I10,12X,3E20.10)
000002       660 FORMAT(//96H   SOURCE          DEGREES OF FRESDOM    SUM OF THE SQUA
                1RES          VARIANCE       VARIANCE RATIO  //)
000002       661 FORMAT(1H(I2,15H(3H X(I2,2H) )))
000002       700 FORMAT(7H RSS = F20.10)
000002       701 FORMAT(13H B-NEW MATRIX//4(6F20.10/))
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000002      702 FORMAT(26H LEAVING OUT VARIABLE NO. I2//13H NEW C-MATRIX//)         054
000002      705 FORMAT(///)                                                          055
000002      720 FORMAT(1H1)                                                          056
000002      721 FORMAT(67X,27H     DEVIATION           O/O   )                       057
000002      722 FORMAT(35H THE AVERAGE ABSOLUTE DEVIATION IS  F10.4,9H PERCENT.)    058
000002     7221 FORMAT (30H ABSOLUTE AVERAGE DEVIATION = ,F20.10,20HCONFIDENCE FAC
               1TOR = ,F20.10)
000002      723 FORMAT(10X,10HINPUT DATA,//,5X,1HY,15X,24HX VALUE(S)-LEFT TO RIGHT  059
               1)                                                                    060
000002      724 FORMAT (8A10)
000002     7241 FORMAT (1X,8A10)
000002      726 FORMAT(//)                                                           061
000002     1327 FORMAT(//21H ANALYSIS OF VARIANCE )                                  062
000002     1628 FORMAT(//24H CONFIDENCE LIMITS OF Y )                                063
000002     4940 FORMAT(19H(26H WITH LIMITS OF,I3,11H PERCENT//))                     064
000002       99 READ 600,INDEX                                                       065
000010          READ 1,NGUESS,MMQ,TYPE
000022          READ 724, (ICELL(I),I=1,8)
000030          READ 601,M,N                                                         066
000040          KKON = 80 + 5 * NGUESS                                               068
000043          ENCODE(33,4940,FORM2)KKON                                            069
000052          PRINT 720
000056          PRINT 7241,(ICELL(I),I=1,8)
000064          PRINT 723$NCTE=0
000071          IF(MMQ.EQ.3HNON)5,599                                                071
000076        5 MG=1  $JMG=1$NOTE=1                                                  072
000100          DO 100 J=1,M                                                         073
000102      100 CALL IDATA(Y(J),X(1,J),NOTE)                                         074
000111          GO TO 883                                                            075
000111      599 MG=2$JMG=0$DO 3 J=1,M                                                076
000115        3 CALL IDATA(Y(J),X(1,J),NOTE)                                         077
000124          N=N-1                                                                078
000125      883 PRINT 726                                                            079
000131          CALL WEIGHT                                                          080
000132      102 DO 104 I=1,N                                                         081
000134          DO 104 J=1,N                                                         082
000135          SUM=0.0                                                              083
000136          DO 103 K=1,M                                                         084
000150      103 SUM=SUM+W(K)*X(J,K)*X(I,K)                                           085
000154      104 A(I,J)=SUM                                                           086
000164          CALL GAUSS3(N,EP,A,C,KER)                                            087
000167          DO 107 I=1,N                                                         088
000171          SUM=0.0                                                              089
000172          DO 106 J=1,M                                                         090
000202      106 SUM=SUM+W(J)*X(I,J)*Y(J)                                             091
000206      107 YX(I)=SUM                                                            092
000212          DO 109 I=1,N                                                         093
000214          SUM=0.0                                                              094
000215          DO 108 K=1,N                                                         095
000225      108 SUM=SUM+YX(K)*C(I,K)                                                 096
000230      109 B(I)=SUM                                                             097
000234          IF(INDEX(8).EQ.2HNO)111,4938                                         098
000241     4938 IF(INDEX(12).EQ.3HYES) 1000,1001                                     099
000246     1000 PRINT 66                                                             100
000252          DO 61 I=1,N                                                          101
000254       61 PRINT 65,(A(I,J),J=1,N)                                              102
000273     1001 IF(INDEX(13).EQ.3HYES) 1002,1003                                     103
000300     1002 PRINT 67                                                             104
```

```
000304          DO 60 I=1,N
000306       60 PRINT 65,(C(I,J),J=1,N)
000325     1003 IF(INDEX(10).EQ.3HYES)42,43
000332       42 DO 44 I=1,N
000334          DO 44 J=1,N
000335          SUM=0.
000336          DO 45 K=1,N
000350       45 SUM=SUM+C(I,K)*A(K,J)
000354       44 CDOT(I,J)=SUM
000364          PRINT 70
000367          DO 71 I=1,N
000371       71 PRINT 65,(CDOT(I,J),J=1,N)
000410       43 IF(INDEX(14).EQ.3HYES) 1004,1005
000415     1004 PRINT 68
000421          PRINT 65,(YX(I),I=1,N)
000430     1005 PRINT 75
000434      111 IF(INDEX(1).EQ.3HYES)7,150
000441        7 DO 8 I=1,N
000443          K=I-JMQ
000444        8 PRINT 605,K,B(I)
000457      150 DO 152 J=1,M
000461          YHAT(J)=0.0
000462          DO 152 I=1,N
000472      152 YHAT(J)=YHAT(J)+X(I,J)*B(I)
000500          SUM1=0.0
000501          DO 153 J=1,M
000512          SUM1=SUM1+Y(J)
000513      153 YBAR=SUM1/FLOAT (M)
000515          SSR=0.0
000515          DIVFR=0.0
000516          DIFF=0.0
000517          DO 154 J=1,M
000527          SSR=SSR+(YHAT(J)-YBAR)**2*W(J)
000532          DIVFR=DIVFR+(Y(J)-YHAT(J))**2*W(J)
000534      154 DIFF =DIFF +(Y(J)-YBAR)**2*W(J)
000540          IF(MMQ.EQ.3HNON)1006,1007
000545     1006 NDR=N-1
000547          GO TO 1008
000550     1007 NDR=N
000552     1008 NDFR=M-N
000554          NTOT = M -1
000555          SMDR=SSR/FLOAT (NDR)
000560          S2=DIVFR/FLOAT (NDFR)
000562          F=SMDR/S2
000563          IF(INDEX(2).EQ.3HYES)151,200
000570      151 PRINT 1327
000574          PRINT 660
000600          PRINT 606,NDR,SSR,SMDR,F,NDFR,DIVFR,S2,NTOT,DIFF
000626          TOTAL = SSR+DIVFR
000630          DEV=DIFF-TOTAL
000631          IF(ABS (DEV)-1.0E-6)160,160,155
000635      155 PRINT 156, DEV
000643      160 PRINT 660
000647          DO 175 I=1,N
000654          NDFR=1
000655          SS2=B(I)*B(I)/C(I,I)
000657          VMS=SS2
000660          F=VMS/S2
```

```
000661          K=I-JMO
000663      175 PRINT 655,K,NDFR,SS2,VMS,F
000704      167 IF(INDEX(11).EQ.3HYES)180,200
000711      180 PRINT 660
000715          READ 607,NO,INDEX(8),INDEX(11)
000727          READ 608,(NSUB(I),I=1,NO)
000736          PRINT 652
000742          DO 157 J=1,NO
000752          K=NSUB(J)+JMO
000753      157 BDOT(J)=B(K)
000756          DO 158 J=1,NO
000757          JJ=NSUB(J)+JMO
000761          DO 158 K=1,NO
000773          KK=NSUB(K)+JMO
000774      158 CDOT(J,K)=C(JJ,KK)
001004          IF(INDEX(8).EQ.3HYES)14,13
001010       14 SWAS=BDOT(1)*BDOT(1)/CDOT(1)
001012          KAZZ=1
001013          F=SWAS/S2
001015          PRINT 37,NSUB(1),KAZZ,SWAS,SWAS,F
001032          DO 15 I=2,NO
001034          CALL GAUSS3(I,EP,CDOT,CINV,KER)
001037          DO 16 JK=1,I
001041          QP(JK)=0.
001042          DO 17 KL=1,I
001052       17 QP(JK)=QP(JK)+CINV(KL,JK)*BDOT(KL)
001055       16 QP(JK)=BDOT(JK)*QP(JK)
001062          S6=0.
001063          DO 18 JK=1,I
001070       18 S6=S6+QP(JK)
001072          VMS=S6-SWAS
001074          SWAS=S6
001075          F=VMS/S2
001076          K=I-1
001101          ENCODE(45,38,FORM)K
001110          PRINT FORM,NSUB(I),(NSUB(J),J=1,K)
001121       15 PRINT 653,KAZZ,VMS,VMS,F
001140       13 CALL GAUSS3(NO,EP,CDOT,CINV,KER)
001144          DO 164JK=1,NO
001146          QP(JK)=0.0
001147          DO 163KL=1,NO
001157      163 QP(JK)=QP(JK)+CINV(KL,JK)*BDOT(KL)
001162      164 QP(JK)=BDOT(JK)*QP(JK)
001167          S6=0.0
001170          DO 165 JK=1,NO
001175      165 S6=QP(JK)+S6
001177          VMS=S6/FLOAT (NO)
001201          F=VMS/S2
001203          ENCODE (18,661,FORM)NO
001213          PRINT FORM,(NSUB(I),I=1,NO)
001222          PRINT 653,NO,S6,VMS,F
001236          GO TO 167
001237      200 IF(INDEX(3).EQ.3HYES)201,250
001244      201 DO 203 I=1,N
001246          DO 203 J=1,N
001256      203 COVAR(I,J)=C(I,J)*S2
001263          PRINT 651
001266          DO 205 I=1,N
```

```
001270       205 PRINT 627,(COVAR(I,J),J=1,N)
001307           PRINT 628
001312           PRINT FORM2
001316       250 IF(INDEX(4).EQ.3HYES)251,300
001323       251 DF=FLOAT (M-N)
001325           I=1
001327       252 IF(TABLE(I,1)-DF)253,255,254
001332       253 I=I+1
001334           GO TO 252
001343       254 IL=I-1
001344           X1=TABLE(IL,NGUESS)
001345           X2=TABLE(I,NGUESS)
001345           Y1=TABLE(IL,1)
001347           Y2=TABLE(I,1)
001351           TE=X1-(Y1-DF)*(X1-X2)/(Y1-Y2)
001357       260 IF(INDEX(4).EQ.3HYES)256,360
001364       255 TE=TABLE(I,NGUESS)
001370       256 DO 257 I=1,N
001372           TLIM=TE*SQRT (S2*C(I,I))
001400           K=I-JKO
001403       257 PRINT 614,K,B(I),TLIM
001417       300 IF(INDEX(5).EQ.3HYES)301,350
001424       301 PRINT 705
001430           DO 305 IJ=1,M
001432           VARI(IJ)=0.0
001433           DO 303 JK=1,N
001435           SL1(JK)=0.0
001436           DO 302 KL=1,N
001451       302 SL1(JK)=X(KL,IJ)*C(KL,JK)+SL1(JK)
001454       303 SL1(JK)=SL1(JK)*X(JK,IJ)
001463           DO 304 JK=1,N
001470       304 VARI(IJ)=VARI(IJ)+SL1(JK)
001473           VA=VARI(IJ)*S2
001475       305 PRINT 615,IJ,VA
001510           PRINT 705
001513       350 IF(INDEX(6).EQ.3HYES)351,400
001520       351 IF(INDEX(5).EQ.3HYES)357,352
001525       352 DO 355 IJ=1,M
001527           VARI(IJ)=0.0
001530           DO 354 JK=1,N
001532           SL1(JK)=0.0
001533           DO 353 KL=1,N
001546       353 SL1(JK)=X(KL,IJ)*C(KL,JK)+SL1(JK)
001551       354 SL1(JK)=SL1(JK)*X(JK,IJ)
001560           DO 355 JK=1,N
001565       355 VARI(IJ)=VARI(IJ)+SL1(JK)
001572       357 IF(INDEX(4).EQ.3HYES)360,251
001577       360 PRINT 1628
001603           PRINT FORM2
001607           SPDY=0.
001610           SDELY=0.
001611           PRINT 721
001614           DO 358 I=1,M
001617           DELY=Y(I)-YHAT(I)
001621           PCY=100.*DELY/Y(I)
001622           SDELY=SDELY+ABS(DELY)
001625           SPDY=SPCY+ABS (PDY)
001630           TLIM=TE*SQRT (S2*VARI(I))
```

```
001636      358 PRINT 616,I,YHAT(I),TLIM,DELY,PDY
001656          IF(TYPE.EQ.7HSEMILOG) 3582,3583
001662     3582 CONTINUE
001662          PRINT 726
001666          AVDELY=SDELY/FLOAT(M)
001670          CONFAC=10.**AVDELY
001674          DO 3581 I=1,M
001676          TLIM=TE*SQRT (S2*VARI(I))
001705          YHATT(I)=10.**YHAT(I)
001711          YUP(I)=10.**(YHAT(I)+TLIM)
001716          YLOW(I)=10.**(YHAT(I)-TLIM)
001723          DELTY(I)=YUP(I)-YLOW(I)
001725     3581 PRINT 6161,I,YHATT(I),YUP(I),YLOW(I),DELTY'I)
001746          PRINT 726
001751          PRINT 7221,AVDELY,CONFAC
001761     3583 CONTINUE
001761          PRINT 726
001765          SPDY=SPDY/FLOAT (M)
001767          PRINT 722,SPDY
001775      400 IF(INDEX(7).EQ.3HYES)401,500
002002      401 R =SSR/DIFF
002004          F=SSR/(S2*FLOAT (N-1))
002010          PRINT 617,R,F
002017      500 IF(INDEX(9).EQ.3HYES)501,2
002024      501 DO 504 I=1,N
002026          DO 503 J=1,N
002027          DO 502 K=1,N
002047      502 CN(J,K)=C(J,K)-(C(J,I)*C(K,I))/C(I,I)
002053          BSTAR(J)=B(J)-(B(I)*C(J,I))/C(I,I)
002063      503 BSS=B(I)*B(I)/C(I,I)
002072          IP=I-JMG
002074          PRINT 702,IP
002101          DO 506 K=1,N
002103      506 PRINT 625,(CN(K,J),J=1,N)
002122          PRINT 705
002125          PRINT 701,(BSTAR(J),J=1,N)
002134          PRINT 705
002140          PRINT 700,BSS
002146      504 PRINT 705
002155        2 READ 600,MMG
002163          IF(MMG.EQ.4HMORE)99,993
002170      993 CONTINUE
002171              END


PROGRAM LENGTH INCLUDING I/O BUFFERS
040302

FUNCTION ASSIGNMENTS

STATEMENT ASSIGNMENTS
1       -  002201    2     -  002156    5     -  000077    7     -  000442
13      -  001141    14    -  001011    37    -  002204    38    -  002211
42      -  000333    43    -  000411    65    -  002220    66    -  002223
67      -  002226    68    -  002233    70    -  002236    75    -  002242
99      -  000003    102   -  000133    111   -  000435    150   -  000460
151     -  000571    155   -  000636    156   -  002250    160   -  000644
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